2016

INDEPENDENT ORIGINATION OF FLORAL ZYGOMORPHY, A PREDICTED ADAPTIVE RESPONSE TO POLLINATORS: DEVELOPMENTAL AND GENETIC MECHANISMS

Ghadeer Bukhari

Wenheng Zhang

INDEPENDENT ORIGINATION OF FLORAL ZYGOMORPHY, A PREDICTED ADAPTIVE RESPONSE TO POLLINATORS: DEVELOPMENTAL AND GENETIC MECHANISMS

A Thesis submitted in partial fulfillment of the requirements for the degree of Master of Science at Virginia Commonwealth University.

by

GHADEER BUKHARI
BACHELOR OF SCIENCE, KING ABDULAZIZ UNIVERSITY, 2009
MASTER OF SCIENCE, VIRGINIA COMMONWEALTH UNIVERSITY, 2016

Director: WENHENG ZHANG, PHD
ASSISTANT PROFESSOR, DEPARTMENT OF BIOLOGY

Codirector: BONNIE L. BROWN, PHD
PROFESSOR AND ASSOCIATE DEPARTMENT CHAIR, DEPARTMENT OF BIOLOGY

Virginia Commonwealth University
Richmond, Virginia
July, 2016

Acknowledgements

It is a great opportunity and pleasure to express my profound gratitude to a number of people to whom I am greatly in debt and appreciate. To my advisor Dr. Wenheng Zhang, a very special thank you for the inspiration, priceless advice, constant guidance, friendship, mentorship, and continuing gifts of wisdom. Thank you for believing in me and for allowing me to grow as a research scientist. I am also very grateful to Dr. Bonnie L. Brown for presenting me with fascinating researches, continuing support, and for sharing the scientific knowledge and experience with me. To my dear parents and siblings, thank you for all of the endless support, encouragement, and caring love you provide for which none of this would have been possible to achieve. To my best friend, dear husband, and great father, Hamza Magsood who remains willing to practically and emotionally supports me as a wife and then a mother of our beloved Eleen, and consistently have faith in me that has been and will be the driving force behind all my achievements. In addition, a great thanks to my committee members: Dr. Wenheng Zhang, Dr. Bonnie L. Brown, Dr. Karen M. Kester, and Dr. Edward L. Boone whom have guided me, supported me, and continue to motivate me to be the best version of myself. In addition, my special appreciation to my great labmates: Dr. Jingbo Zhang, Farahnoz Khojayori, Joon Kim, and Ao Gao for their friendship and support in providing assistance to complete my study.

Table of Contents

List of Tables

List of Figures

List of Abbreviations

MAB: Medially abaxial petal initiation.

MAD: Medially adaxial petal initiation.

CV: Central ventral petal.

CD: Central dorsal petal.

ML: Maximum likelihood.

MrBayers: Baysian

bp: base pair

Mbp: million base pairs

RPKM: Reads per kilobase of transcript per million mapped reads

Abstract

INDEPENDENT ORIGINATION OF FLORAL ZYGOMORPHY, A PREDICTED ADAPTIVE RESPONSE TO POLLINATORS: DEVELOPMENTAL AND GENETIC MECHANISMS

By Ghadeer Bukhari, B.S.

A thesis submitted in partial fulfillment of the requirements for the degree of Master of Science in Biology at Virginia Commonwealth University.

Virginia Commonwealth University, 2016

Major Advisor: Wenheng Zhang
Assistant Professor, Department of Biology

Coadvisor: Bonnie L. Brown
Professor and Associate Department Chair, Department of Biology

Observations of floral development indicate that floral organ initiation in pentapetalous flowers more commonly results in a medially positioned abaxial petal (MAB) than in a medially positioned adaxial petal (MAD), where the medial plane is defined by the stem and the bract during early floral development. It was proposed that the

dominant MAB petal initiation might impose a developmental constraint that leads to the evolution of limited patterns of floral zygomorphy in Asteridae, a family in which the floral zygomorphy develops along the medial plane and results in a central ventral (CV) petal in mature flowers. Here, I investigate whether the pattern of floral organ initiation may limit patterns of floral zygomorphy to evolve in pentapetalous angiosperms. I analyzed floral diagrams representing 405 species in 330 genera of pentapetalous angiosperms to reconstruct the evolution of floral organ initiation and the evolution of developmental processes that give rise to floral zygomorphy on a phylogenetic framework. Results indicate that MAB petal initiation is the most common; it occupies 86.2% of diversity and represents the ancestral state of floral organ initiation in pentapetalous angiosperms. The MAD petal initiation evolved 28 times independently from the ancestral MAB petal initiation. Among the 34 independent originations of floral zygomorphy, 76.5% of these clades represent MAB petal initiation, among which only 47% of the clades result a CV petal in mature flowers. The discrepancy is explained by the existence of developmental processes that result in floral zygomorphy along oblique planes of floral symmetry in addition to along the medial plane. Findings suggest that although the early floral organ initiation plays a constraining role to the evolution of patterns of floral zygomorphy, the constraint diverges along phylogenetically distantly related groups that allow the independent originations of floral zygomorphy through distinct development processes in pentapetalous angiosperms.

In additional study, the butterfly-like flowers of *Schizanthus* are adapted to pollination by bees, hummingbirds, and moths. I investigated the genetic basis of the zygomorphic corolla, for which development is key to the explosive pollen release mechanism found in the species of *Schizanthus* adapted to bee pollinators. I examined differential gene expression profiles across the zygomorphic corolla of *Schizanthus pinnatus*, a bee-pollinated species, by analyzing RNA transcriptome sequencing (RNA-seq). Data indicated that *CYC2* is not expressed in the zygomorphic corolla of *Sc. pinnatus*, suggesting *CYC2* is not involved in the development of floral zygomorphy in *Schizanthus* (Solanaceae). The data also indicated that a number of genes are differentially expressed across the corolla.

Chapter One

Introduction

Most of the flowers can be classified as either actinomorphic (radially symmetrical, polysymmetric) with multiple planes of floral symmetry, or as zygomorphic (bilaterally symmetrical, monosymmetric) with a single plane of floral symmetry (Endress, 2001). Paleontological studies indicate the earliest fossils of flowers are actinomorphic, which are found from the Upper Cretaceous (Turonian) period, while the zygomorphic flowered species was found until 30-40 millions years afterwards (Crane, Friis, & Pedersen, 1995; Crepet, 1996). Phylogenetic studies have also shown that floral zygomorphy had evolved independently numerous times from ancestors with actinomorphic flowers (Bukhari, Zhang, & Zhang, 2015; Reyes, Nadot, & Sauquet, 2015). Zygomorphic flowers are the traits associated with the greatly divergent clades such as, Orchidales, Lamiales, Dipsacales, Fabaceae, and Asteraceae (Endress, 2001). Studies have shown that the floral zygomorphy is preferred by some pollinators with symmetry preference such as the bees (Giurfa, Dafni, & Neal, 1999).

Recent advances in developmental genetics start to reveal the molecular mechanisms underlying the evolution of floral zygomorphy in angiosperms (Busch, Horn, & Zachgo, 2014). While the examination of the evolutionary patterns and the molecular mechanisms to generate floral zygomorphy has been the focus of most researches, how the development may influence the morphological evolution of floral zygomorphy is rarely investigated. The only investigation in Asteridae by Donoghue, Ree, and Baum (1998) attempted to use the developmental process to explain the law of zygomorphic floral patterns in this clade. In Asteridae, the zygomorphic flowered species possess three major types of floral zygomorphy, which are described as 2:3, 4:1, and 0:5 based on the number of the petals located on the dorsal versus ventral side of the flower (Donoghue et al., 1998). Among the three major zygomorphic types, the 2:3 is most common, while 0:5 is the rarest. Donoghue et al. (1998) proposed that the limited types of floral zygomorphy observed in Asteridae is rooted in the conserved pattern of floral organ initiation of their flowers, most of which possess a medially positioned abaxial (MAB) petal (Fig. 1 A) (Donoghue et al., 1998; Eichler, 1875-78). When zygomorphy develops along the medial plane of floral symmetry, which is defined by the stem and the bract, the three major types of floral zygomorphy in Asteridae are the only possible variations, which exhibit the central ventral (CV) petal in mature flowers (Fig. 1C). However, Donoghue et al. (1998) did mention rare exceptions in Asteridae, of which species possess a medially adaxial (MAD) petal at early stage of flower development (Fig. 1B). For example, the early MAD floral development of Rhododendron (Ericaceae) associates with its 3:2 type of floral zygomorphy, which exhibits the central dorsal (CD) petal at later stage of floral development (Fig. 1D). The study in Asteridae suggests the dominant MAB petal initiation of floral development found in this group might play a role to constrain the types of floral zygomorphy to evolve.

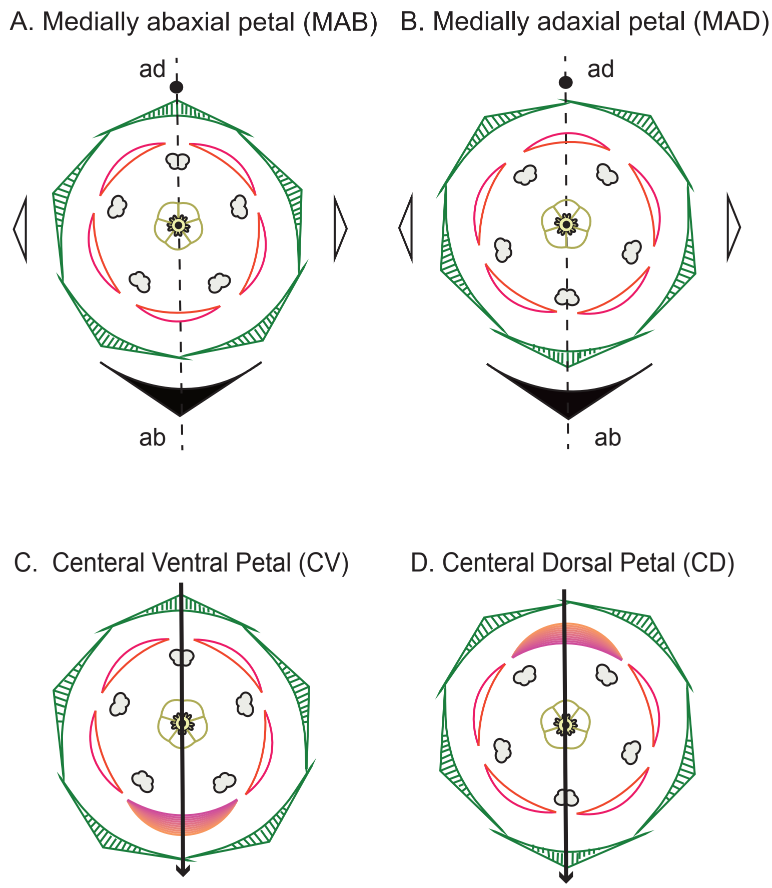


Figure 1. Floral initiation and petal arrangement of floral zygomorphy in pentapetalous angiosperms. There are two major orientations of floral initiation in pentapetalous angiosperms. It is called (A) medially positioned abaxial petal initiation (MAB) if the medially positioned petal locates at the abaxial side of the flower close to the bract, or (B) medially positioned adaxial petal initiation (MAD) if the medially positioned petal locates at the adaxial side of the flower close to the axis. There are two major ways of petal arrangement for the pentamerous zygomorphic flowers. They are (C) Central ventral petal (CV) if the medially positioned petal located at the abaxial

position, or (D) Central dorsal petal (CD) if the medially positioned petal located at the adaxial position. The stem axis is indicated by solid circle. The bract is indicated as solid leaf-like structure opposed to the axis. The bractlets are indicated by open leaf-like structures. The four whorls of the flower from outside to the center are calyx, corolla, androecium, and gynoecium. The pentamerous syncarpous gynoecium is hypothetical for the illustration.

In this study, we investigate if the floral organ initiation has imposed constraint in the development of the pattern of floral zygomorphy across pentapetalous angiosperms. We examine the evolution of floral initiation patterns, and whether the patterns of floral initiation (MAB vs. MAD petal initiations) would limit how the patterns of floral zygomorphy (CV vs. CD petal patterns) to develop in the mature flower. That is to test if the MAB petal initiation would commonly result floral zygomorphy that displays the CV petal, while if the MAD petal initiation would commonly result floral zygomorphy that displays the CD petal. Since previous observations indicate that floral organ initiation in pentapetalous flowers are most commonly with a MAB petal initiation, we would like to test the hypothesis that there would be more of floral zygomorphy with CV petal among zygomorphic flowered clades.

We analyzed floral diagrams to include 405 species in 330 genera representing all the major lineages of pentapetalous angiosperms. We extracted four floral characters from the floral diagrams including patterns of floral organ initiation (MAB vs. MAD), patterns of floral symmetry (actinomorphy vs. zygomorphy), patterns of floral zygomorphy (CD vs. CD), and developmental processes to establish floral zygomorphy (see Results and Dissection). Then, we reconstructed a phylogenetic tree for the 405 sampled species to carry out the ancestral state reconstruction for the four characters. Our findings suggest that although the early floral organ initiation plays a role as constraint to the evolution of displays of floral zygomorphy, the constraint diverges along phylogenetically distantly related groups that allow the independent originations of floral zygomorphy through distinct development processes in pentapetalous angiosperms. We argue that identification and systematically studies of diverse developmental processes that give rise to floral zygomorphy will help us to understand how these developmental processes link with the genetic pathways responsible for the independent origins of floral zygomorphy.

Materials and Methods

Characterizing traits from floral diagram:

Flower diagrams from the works of three botanists including Eichler (1875-1878), Engler (1887–1915), and De Craene (2010) were analyzed (Fig. 2) (De Craene, 2010; Eichler, 1875-78; A. Engler & K Pranrl, 1895; Engler & Pranrl, 1897a, 1897b; Engler & Pranrl, 1889a, 1889b, 1889c, 1891a, 1891b, 1893, 1894; A. Engler & K. Pranrl, 1895; Engler & Pranrl, 1897a, 1897b, 1898). We collected the character states of four floral traits for the pentapetalous angiosperms (Appendix table 1). These floral traits include: 1) Patterns of floral organ initiation, which describes the relative position of the medially positioned petal at the early stage of floral development, that is MAB versus MAD petal initiation (Fig. 1 A and B). For the MAB petal initiation, the medially positioned petal initiates from the abaxial position of the flower close to the bract. Vise versa, for the MAD petal initiation, the medially positioned petal initiates from the adaxial position of the flower close to the axis (Fig.1 A and B). 2) Patterns of floral symmetry, which describes whether the flower is actinomorphic or zygomorphic (Fig. 1 C and D). 3) Displays of floral zygomorphy, which describes whether the zygomorphic flower has a CV petal or a CD petal in the mature flower (Fig.1 C and D). The patterns of CD or CV can be determined based on whether the central petal penetrated by the plane of floral symmetry locates on the dorsal side or ventral side of the mature flower. And 4) developmental processes to establish floral zygomorphy, which describes by three parameters, which are the pattern of floral initiation, the angle of floral symmetrical plane relative to the medial plane, and the displays of floral zygomorphy (Fig. 3, also see the Results and Discussion). These three parameters are sufficient to describe unique developmental processes to give rise to floral zygomorphy. The oblique plane of floral symmetry can be determined based on the dotted line illustrated across the floral diagram.

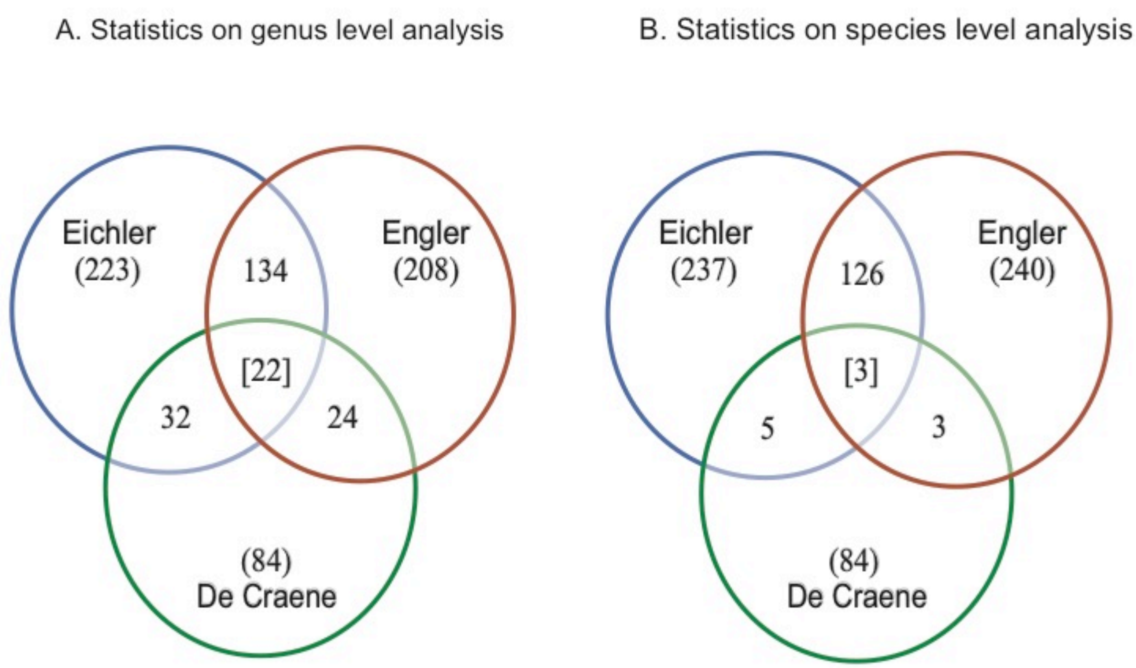


Figure 2. Data statistics. (A) Statistics on genus level diversity. (B) Statistics on species level diversity. The taxa sampling is based on literatures from Eichler 1875-1878, Engler 1887–1915, and Ronse de Craene, 2010 (De Craene, 2010; Eichler, 1875-78; A. Engler & K Pranrl, 1895; Engler & Pranrl, 1897a, 1897b; Engler & Pranrl, 1889a, 1889b, 1889c, 1891a, 1891b, 1893, 1894; A. Engler & K. Pranrl, 1895; Engler & Pranrl, 1897a, 1897b, 1898). The numbers inside the circled brackets indicate the

unique genera/species covered in each work. The numbers inside the squared brackets indicate the genus/species overlapped in all works. The numbers without brackets indicate the shared genus/species between two works.

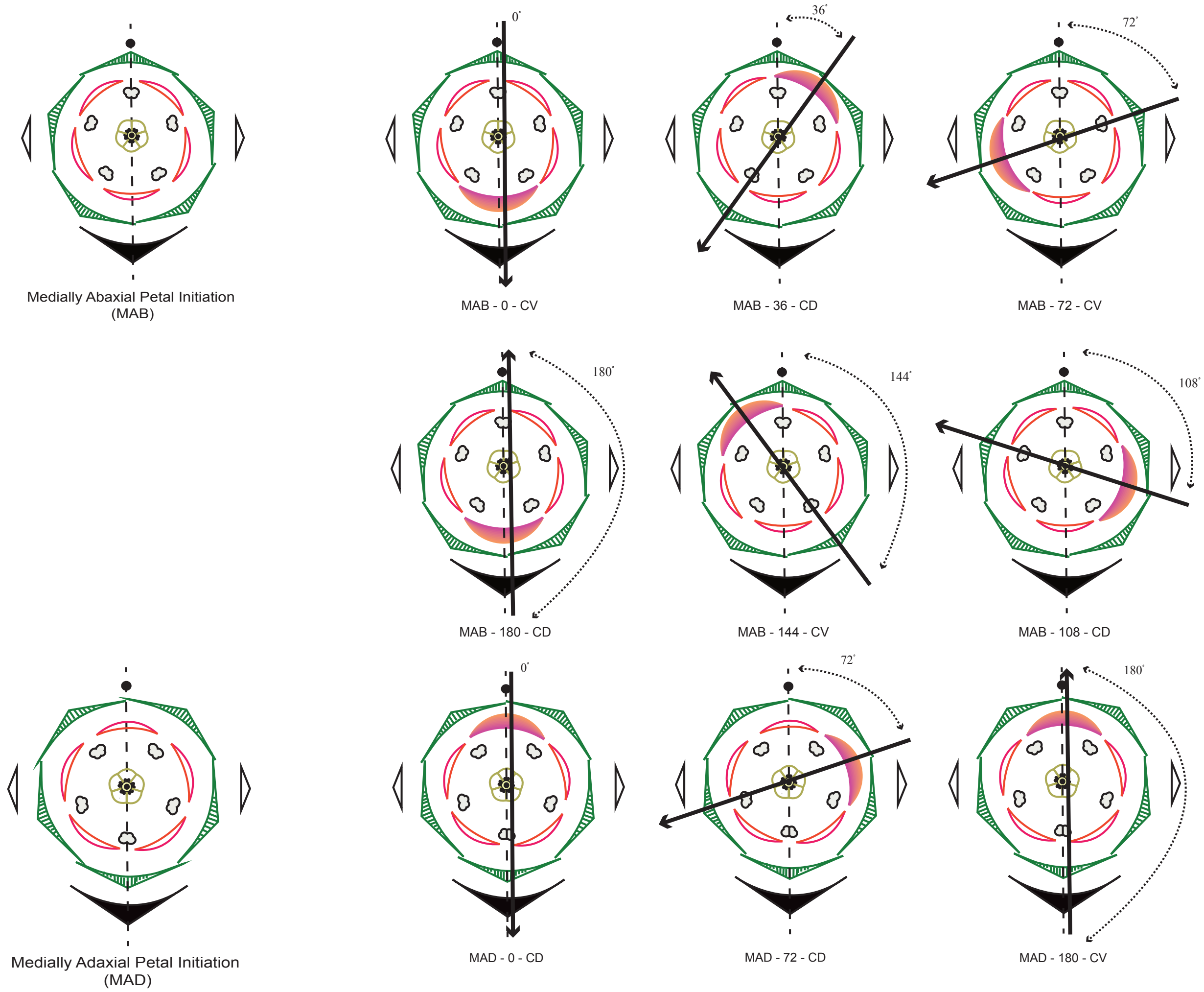


Figure 3. Distinct mechanisms to develop floral zygomorphy from MAB floral initiation. The developmental mechanisms are described based on three parameters: 1) floral initiation (MAB vs. MAD). 2) the angle of the plane of floral symmetry (n degrees) relative to the medially plane. 3) patterns of floral zygomrophy (CV vs. CD); The doted line indicates a medially plane of floral symmetry. The solid line with arrow indicates the plane of floral symmetry established at the later stage of floral

development. All of these patterns had been observed based on our dataset for the MAB petal initiation, and only three of the patterns, which are MAD-F-0, MAD-F-72, and MAD-L-180, had been observed based on the MAD petal initiation on our dataset. Note, symmetrical planes with an angle over 180 degrees were considered as the same as the one they mirrored the ones displayed.

Phylogenetic reconstruction of pentapetalous angiosperms:

The full analysis included 405 unique taxa representing the major lineages of pentapetalous angiospmers. To reconstruct the phylogenetic relationships, the analyses were carried out in the following four steps. First, the taxon names and their taxonomic treatment were checked and updated based on current taxonomic treatment referencing the Angiosperm phylogeny website (APWeb) (Stevens, 2001 onwards), The Plant List website ("The Plant List," 2010), and The International Plant Names Index (IPNI) ("The International Plant Names Index ", 2012) (Appendix table. 1). Second, phylogenetic relationships were extracted to include these taxa using Phylomatic v3 (Webb & Donoghue, 2004). The topology obtained, however, lacks resolutions at the lower taxonomic levels. Third, to fully resolve the tree, we either based on the recent publications on molecular phylogeny of the clade of interests (see references in appendix table. 2); or carried out phylogenetic analyses using DNA sequencing data from Genbank (Sayers et al., 2009). By using the DNA sequencing data, we reconstructed 11 phylogenetic trees on genus and species levels using the Maximum likelihood and Bayesian approaches (Appendix fig. 4). Finally, the fully resolved phylogeny was prepared manually using software Mesquite (Maddison & Maddison, 2015).

Reconstructing ancestral states for floral traits:

The ancestral character state reconstruction of the four floral traits was carried out using parsimony optimization approach implemented in Mesquite (Maddison & Maddison, 2015). The four floral traits, i.e., patterns of floral initiation, patterns of floral symmetry, patterns of floral zygomorphy, and developmental processes to establish floral zygomorphy, were separately analyzed on the fully resolved tree (Appendix tables 1-3 and Figs. 5-8).

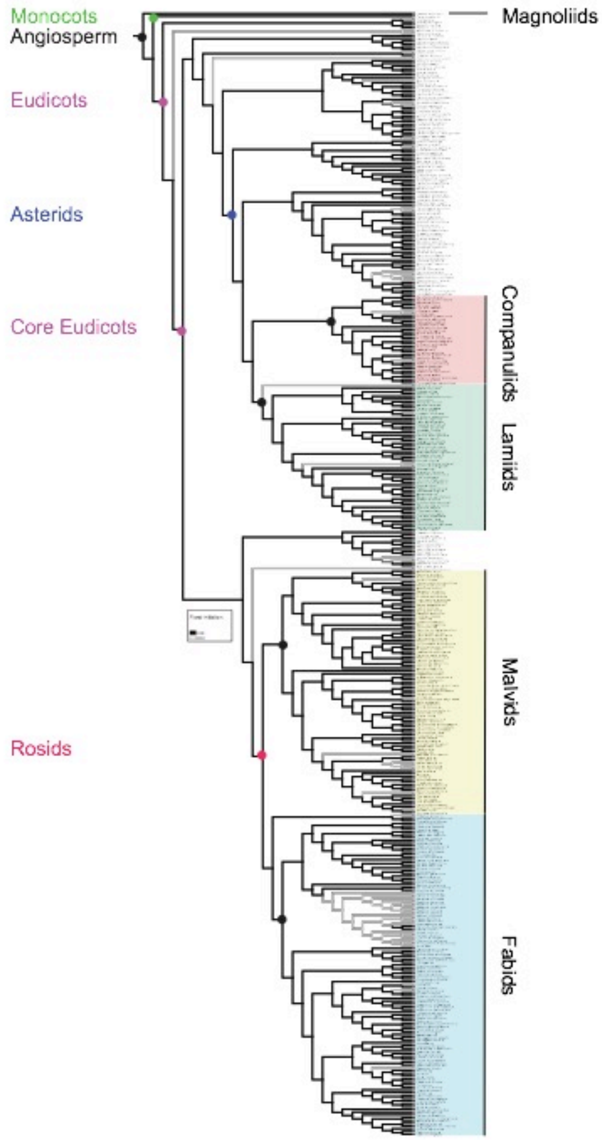


Figure 5. Ancestral state reconstruction of floral initiation. The black color indicates the medially abaxial petal initiation (MAB), and the white color indicates the medially adaxial petal initiation (MAD).

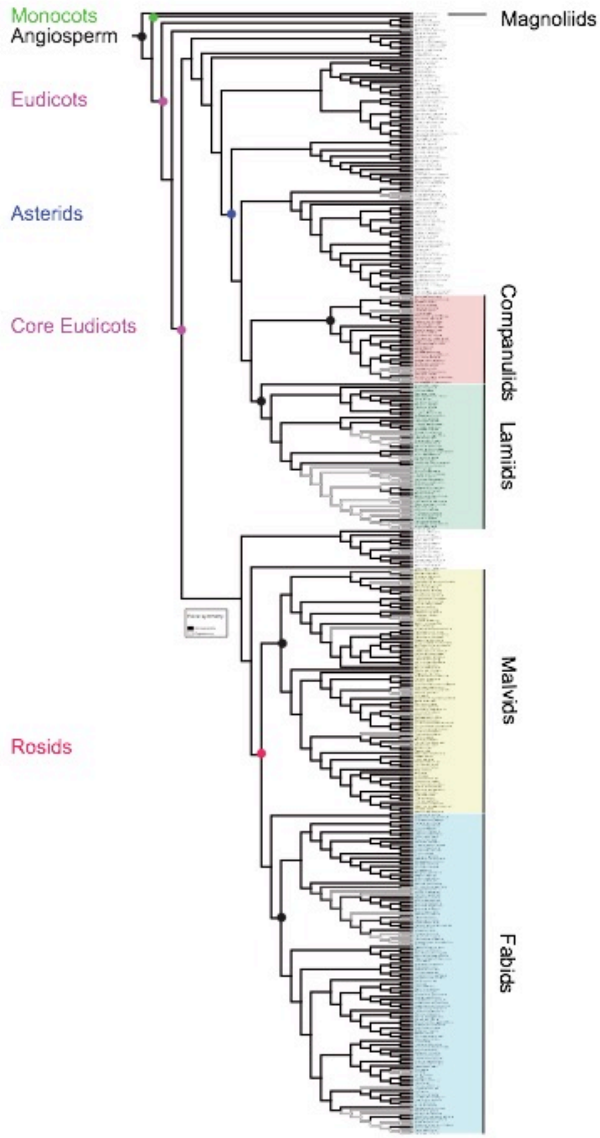


Figure 6. Ancestral state reconstruction of floral symmetry. The black color indicates floral Actinomorphy, and the white color indicates floral Zygomorphy.

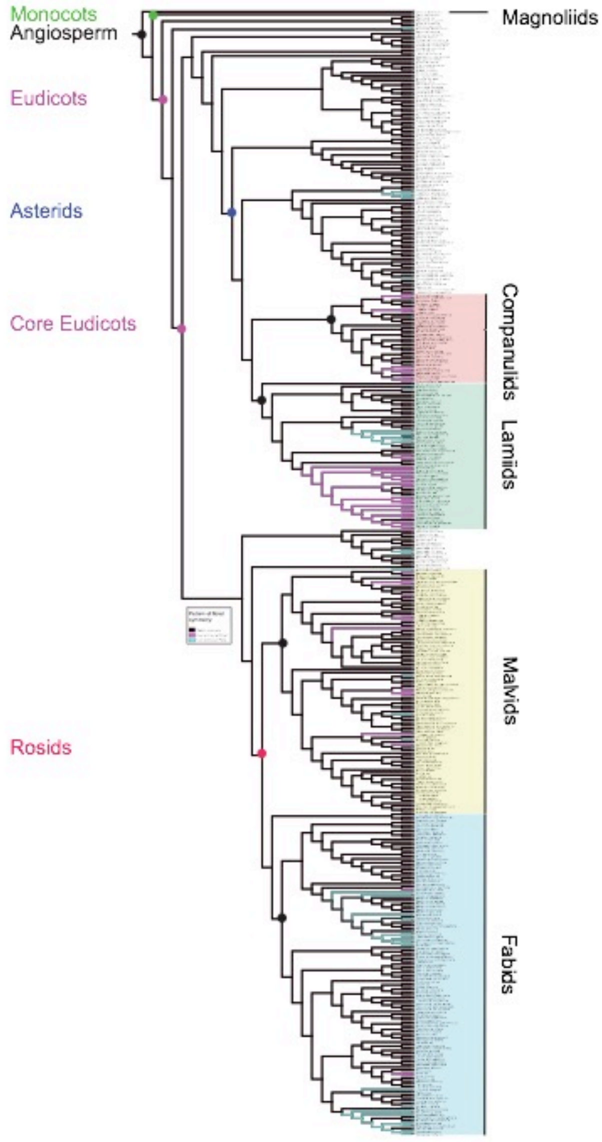


Figure 7. Ancestral state reconstruction of patterns of floral symmetry. The black color indicates the radially symmetrical flowers (R), and the pink color indicates the flowers with a central ventral petal (CV), and the blue color indicates the flowers with a central dorsal petal (CD).

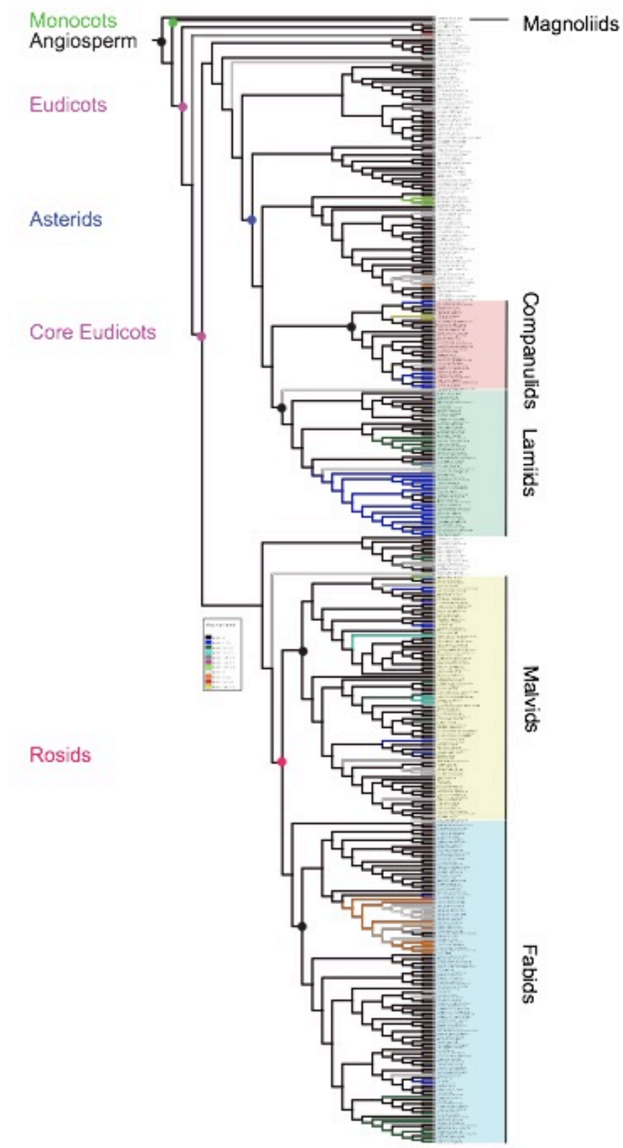


Figure 8. Ancestral state reconstruction of development processes to establish floral zygomorphy. The floral actinomorphy is labeled as: (1) MAB-A, and (2) MAD-A. The first part of the code indicates the floral initiation (MAB vs. MAD), and the second part indicates floral actinomorphy. We also recognize nine development processes that give arise to the floral zygomorphy: (1) MAB-0-CV, (2) MAB-36-CD, (3) MAB-72-CV, (4) MAB-108-CD, (5) MAB-144-CV, (6) MAB-180-CD, (7)

MAD-0-CD, (8) MAD-72-CD (9) MAD-180-CV. The first part of the code indicates the floral orientation (MAB vs. MAD), the second part of the code indicates the degree of the plane of symmetry relative to the medially plane, and the last part indicates the patterns of floral symmetry (CV vs. CD).

Results and Discussion

<u>*Diversity of pentapetalous angiosperms:*</u>

Results show that pentapetalous angiosperms are found in almost all the representatives of flowering plants. The majority of pentapetalous flowers are found in Eudicots including basal and core eudicots. One represents monocots, *Paris polyphylla* (Melanthiaceae), which is illustrated as pentapetalous flowers in a floral diagram (De Craene, 2010) even though individual plants may have flowers with 4 or 6 petals (Stevens, 2001 onwards), and one represents magnoliids, *Canella winterana* (Canellaceae) (Fig. 8, Appendix table. 1). Recent systematic studies have proposed a Pentapetale clade, which includes all core eudicots other than Gunnerales (De Craene, Soltis, & Soltis, 2003; D. E. Soltis et al., 2003; P. S. Soltis & Soltis, 2004). Based on the datasets generated in this study, pentapetalous flowers presented outside the Pentapetale clade, which suggests independent origination of the pentapetalous state outside core eudicots. The eudicots with approximately 75% of the flowering plant species represent the major diversity of pentapetalous angiosperms (Nepokroeff & Zimmer, 2002).

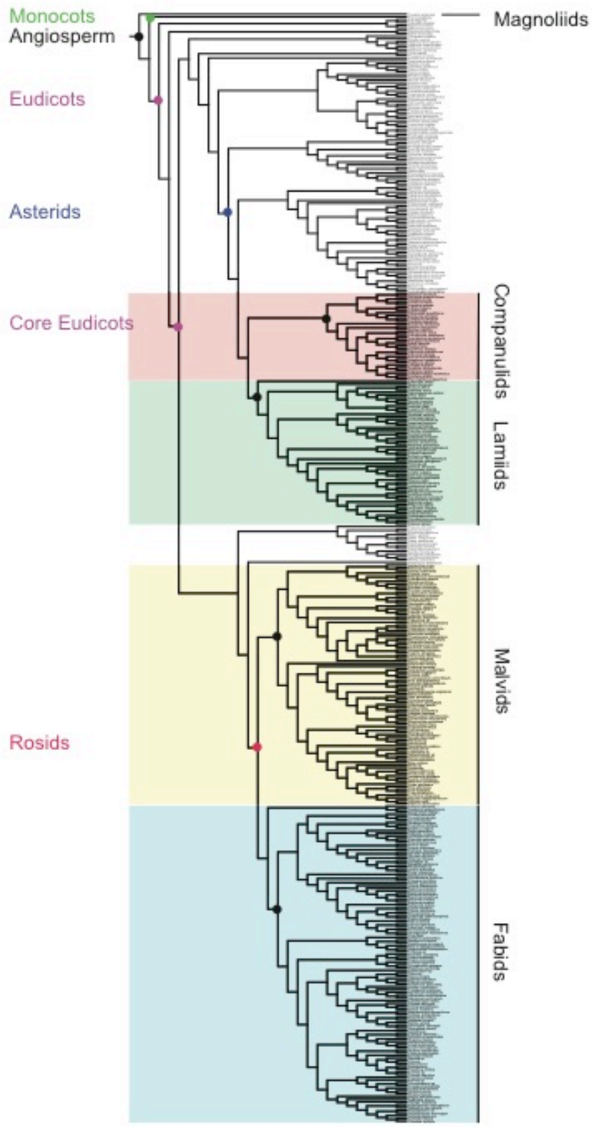


Figure 9. Fully resolved phylogeny tree of angiosperms in the dataset. Major clades (indicated by colored shades) of pentapetalous flowring plants of 405 taxa are included in the fully resolved phylogeney tree.

Evolution of MAB and MAD petal initiation:

Analyses indicate that MAB petal initiation is dominant, which represents 86.2% of the pentapetalous angiosperms (Appendix table. 1). The ancestral state reconstruction indicates that the MAB petal initiation is the ancestral state, while the MAD petal initiation independently evolved 28 times from the ancestors with MAB petal initiation (Fig. 5). We show that MAB petal initiation is most common in Pentapetale clade and pentapetalous flowers outside Pentapetale clade.

Distinct developmental processes to give rise to floral zygomorphy:

Through study of the floral diagrams, it is clear that floral zygomorphy can develop along oblique planes of floral symmetry other than along the medial plane for either the MAB or MAD petal initiations. If the symmetrical plane develops along and parallel to the medial plane, 0° will describe this developmental process. If the symmetrical plane develops along one of the five oblique planes that forms an angle with the medial plane, the angles 36°, 72°, 108°, 144°, or 180° will describe these developmental processes (Appendix table. 1, Fig. 8). The symmetrical planes with an angle over 180° that form mirrored images of these described processes were considered as the same developmental processes (Fig. 3). In this dataset, all six possible symmetrical planes were observed in the zygomorphy flowers with MAB petal initiation, and three possible symmetrical planes (0°, 72°, 180°) were observed in the zygomorphy flowers with MAD petal initiation (Fig. 3). Nine unique developmental processes to give rise to floral zygomorphy were identified from this dataset (Fig. 3).

It turns out that MAB petal initiation contributed to the most of the zygomorphic flowered clades [26 (76.5%) out of the 34 clades], and MAB petal initiation is the major contributor and contributes equally to both the CV petal zygomorphy (15 out of 16 clades) and CD petal zygomorphy (11 out of 18 clades) (Fig. 10, and Table. 4 and 5). The finding of existence of multiple mechanisms to develop floral zygomorphy through diverse planes helps us to understand how both CV and CD petal zygomorphy can be achieved through the MAB petal initiation.

Considering the developmental processes allowed us to identify detailed evolutionary transitions during floral symmetry evolution. When we treated floral zygomorphy as a single trait, we found floral zygomorphy independently evolved 29 times (Fig. 6). However, when we considerate the different development processes we found that floral zygomorphy evolved at least 34 times (Fig. 7 and 8). For example, floral zygomorphy of *Polygala myrtifolia* (Polygalaceae) and Fabaceae was thought to share common ancestry if only consider floral zygomorphy (Fig. 6). However, based on the developmental processes, we know floral zygomorphy development through distinct processes in Polygalaceae (MAB-0-CV) and in Fabaceae (MAD-0-CD) (Fig. 7 and 8).

Some of the mechanisms may represent a twist of the same mechanism. *Melianthus major* and *Bersama maxima* are zygomorphic species in Melianthaceae. Based on our system of description, *Melianthus major* is MAB-180-CD, and *Bersama maxima* is MAB-0-CV. It is likely the two represent the similar mechanism, the 180 is due to the physical movement of the floral orientation due to the imbalanced weight of the flower and its response to gravity.

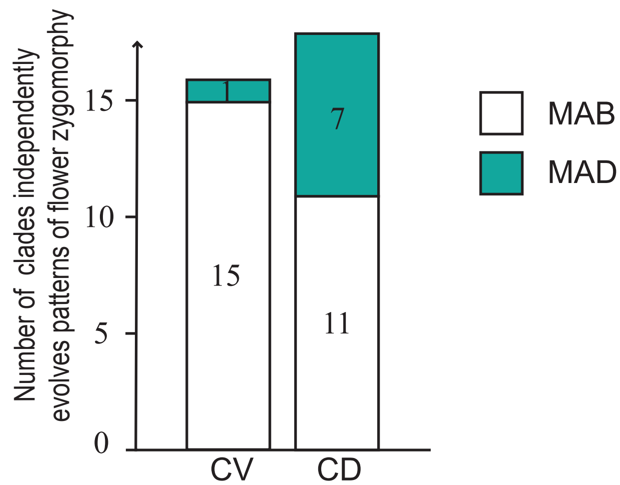


Figure 10. Frequencies of floral zygomorphy with CV and CD patterns evolved from MAB and MAD floral initiations. Among the 16 CV patterns identified, one develops from MAD floral initiation and 15 from MAB floral initiation. Among the 18 CD patterns identified, 7 develop from MAD floral initiation and 11 from MAB floral initiation. Both MAB and MAD floral initiations contribute to both the zygomorphic floral patterns, with MAB being the major contributor to most zygomorphic flowered clades analyzed in this dataset.

Table 4. Developmental processes associated with clades of floral zygomorphy with CV patterns of symmetry.

Clade	Order with CV	Species with CV	CV Pattern
1	Asterales	*Lobelia fulgens, Lobelia tupa*	MAD-180-CV
2	Asterales	*Stylidium adnatum, Stylidium graminifolium*	MAB-0-CV
3	Dipsacales	*Scabiosa atropurpurea, Lonicera giraldii, Pterocephalus palaestinus, Valeriana repens, Linnaea borealis*	MAB-0-CV
4	Boraginales	*Echium hierrense*	MAB-144-CV
	Boraginales	*Echium vulgare*	MAB-72-CV
5	Lamiales	*Acanthus mollis, Bignonia unguis, Clerodendrum petasites, Diascia vigilis, Eranthemum nervosum, Globularia nudicaulis, Gratiola officinalis, Lamium album, Lathraea squamaria, Linaria vulgaris, Odontonema strictum, Orobanche flava, Penstemon fruticosus, Pinguicula alpina, Salvia officinalis, Sinningia sp, Utricularia vulgaris, Veronica chamaedrys, Hebenstretia dentata*	MAB-0-CV
6	Geraniales	*Bersama maxima*	MAB-0-CV
7	Geraniales	*Pelargonium abrotanifolium, Pelargonium zonale*	MAB-0-CV
8	Geraniales	*Erodium leucanthum*	MAB-0-CV
9	Myrtales	*Lopezia racemosa, Lopezia sp*	MAB-0-CV
10	Myrtales	*Salvertia convallariaeodora*	MAB-72-CV
11	Sapindales	*Aesculus hippocastanum*	MAB-72-CV
12	Sapindales	*Serjania glabrata, Urvillea sp*	MAB-72-CV
13	Brassicales	*Tropaeolum majus*	MAB-0-CV
14	Brassicales	*Sesamoides clusii*	MAB-0-CV
15	Fabales	*Polygala myrtifolia*	MAB-0-CV
16	Malpighiales	*Viola sp, Viola tricolor*	MAB-0-CV

Table 5. Developmental processes associated with clades of floral zygomorphy with CD patterns of symmetry.

Clade	Order with CD	Species with CD	CD patterns
1	Sabiales	*Meliosma dilleniifolia*	MAD-72-CD
2	Ericales	*Rhododendron luteum*	MAD-0-CD
3	Fabales	*Afzelia quanzensis*	MAD-0-CD
4	Fabales	*Cercis siliquastrum*	MAD-0-CD
5	Fabales	*Amherstia nobilis*	MAD-0-CD
6	Fabales	*Cassia caroliniana*	MAD-0-CD
7	Fabales	*Chorizema cordatum, Cytisus laburnum, Laburnum vulgare, Strongylodon macrobotrys, Vicia faba*	MAD-0-CD
8	Malpighiales	*Camarea hirsuta, Camarea triphylla, Stigmaphyllon multilobum, Stigmaphyllon sp, Hiptage madablota, Galphimia glauca*	MAB-36-CD
9	Malpighiales	*Hirtella triandra*	MAB-36-CD
10	Malpighiales	*Trigonia simplex*	MAB-36-CD
11	Brassicales	*Moringa oleifera*	MAB-108-CD
12	Sapindales	*Ertela trifolia*	MAB-36-CD
13	Sapindales	*Anacardium occidentale*	MAB-36-CD
14	Geraniales	*Melianthus major*	MAB-180-CD
15	Saxifragales	*Saxifraga fortunei, Saxifraga stolonifera*	MAB-36-CD
16	Solanales	*Hyoscyamus albus, Petunia nyctaginiflora, Salpiglossis sinuata, Schizanthus retusus*	MAB-36-CD
17	Gentianales	*Exacum affine*	MAB-108-CD
18	Ericales	*Impatiens glandulifera, Impatiens platypetala, Impatiens scabrida*	MAB-180-CD

Development constraint process to give rise to floral zygomorphy, phylogenetic distance diverges constraints:

In three clades, we found independent evolution of floral zygomorphy always associating with the use of the same pattern of developmental process. First, in Malpighiales, the CD petal zygomorphy has evolved independently three times in three families; i.e., Malpighiaceae, Chrysobalanaceae, and Trigoniaceae. Interestingly, each time floral zygomorphy evolved by using the same pattern of developmental process, an oblique plane of symmetry with a MAB-36°-CD process (Fig. 3B). Second, in Fabaceae, the conserved pattern of legume flowers are homoplastic based on both the phylogenetic reconstruction and the ontogenetic study of the basal (paraphyletic Caesalpinioid) legumes (Bruneau, Klitgaard, Fougere-Danezan, & Tucker, 2005). This suggests the MAD-0°-CD process identified for the majority of the zygomorphic species in Fabaceae has been independently evolved at least five times (Fig. 3G). Third, our analysis of Asteridea further illustrated Donoghue's point, which shows three independent origins of floral zygomorphy through the MAB-0°-CV process in Stylidiaceae, Dipsacales, and Lamiales. However, six clades of Asteridea were identified to have independently evolved using other processes. Among the independently evolved clades are *Impatiens* (MAB-180°-CD), *Rhododendron* (MAD-0°-CD), *Lobelia* (MAD-180°-CV), *Exacum affine* (MAB-108°-CD), *Echium hierrense* and *E. vulgare* (MAB-144°-CV and MAB-72°-CV, respectively), and Solanaceae (MAB-36°-CD). Although they comprise a minority compared with the diversity of other clades in Asteridea, they represent phylogenetically independent events in the evolution of floral zygomorphy. Distinct processes giving rise to floral zygomorphy in Asteridea can be explained by phylogenetic distance in some cases, such as *Impatiens* and *Rododendron* of

Ericales, and relaxed genetic and developmental processes due to genome duplication, such as in Solanaceae (Aversano et al., 2015; Bombarely et al., 2016; P. G. S. Consortium, 2011; T. G. Consortium, 2012; Qin et al., 2014).

Developmental processes and genetic mechanisms underlying floral symmetry evolution:

Recent advances discovered *CYCLOIDEA*- (*CYC2*-) like genes dependent mechanism for controlling the development of floral zygomorphy. The *CYC2*-dependent mechanism was independently recruited to establish floral zygomorphy in several core eudicots lineages, including Asterales, Brassicales, Dipsacales, Fabales, Lamiales, and Malpighiales (Citerne, Jabbour, Nadot, & Damerval, 2010; Hileman, 2014a, 2014b; Preston, Martinez, & Hileman, 2011; Rosin & Kramer, 2009). Interestingly, all zygomorphy clades examined to date established the floral zygomorphy along the medial plane. The only exception in Malpighiaceae, however initiate the floral zygomorphy along the medial plane and then shifted to an oblique plane with a 36 degree. We are interested in testing to what extent different developmental process will associate with different genetic mechanisms. Therefore, we may predict that more diversified genetic mechanisms; i.e., *CYC2*-independent mechanisms, may also involved in other processes of floral zygomorphy development in core eudicots. This study is the first attempt to clarify the developmental process to give rise to zygomorphic flowers across pentapetalous angiosperms, which provides the first step towards deeper understanding of the role of development in evolution of floral zygomorphy.

Displays of floral zygomorphy and pollinator selection:

Floral zygomorphy was thought to be established through pollinator selections (Gomez, Perfectti, & Camacho, 2006; Ushimaru, Dohzono, Takami, & Hyodo, 2009). Among the floral zygomorphic clades, we observed the approximately equal numbers of clades to evolve CV petal zygomorphy that may function as a landing platform and CD petal zygomorphy that may function as a flag (Fig. 11). Recent studies indicate that the change in pollination niches is directly associated with changes in floral morphology, which indicates the patterns of flower symmetry are indeed distinguished and preferred by certain pollinators (Gomez et al., 2006; Gomez, Perfectti, & Lorite, 2015). However, It is unclear if pollinator selection plays a role in determining which pattern of floral zygomorphy to evolve. Our data suggest that there are equal appreciations by pollinator to evolve both patterns of floral zygomorphy through distinct development processes (Fig. 11).

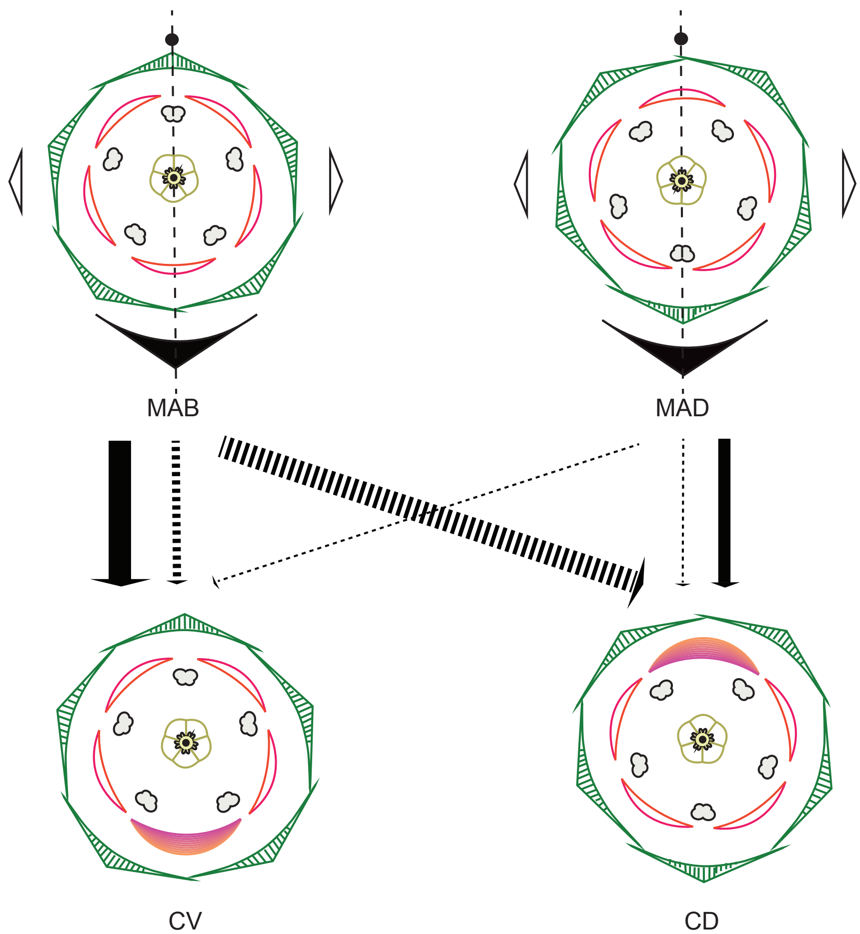


Figure 11. Summary of the developmental processes giving arise to floral zygomorphy. The contributions of MAB, and MAD to establish the CV and CD patterns of floral zygomorphy were illustrated. The MAB contributes to most of the independent origination of floral zygomorphy while the MAD contributes less. The solid lined arrows indicate the floral pattern results in medially plane of symmetry. The doted lined arrows indicate the floral pattern achieved through oblique plane of symmetry. The thickness of the line represents the ratio of their contribution.

Conclusion:

The pattern of petal initiation does not constrain the pattern of floral zygomorphy to evolve. In addition, the phylogenetically closely related species tend to evolve floral zygomorphy using the same developmental process, but the phylogentic distance and the genome duplication may diverge developmental processes.

Chapter Two

Introduction

Differential representation of flower morphology can be the result of selection by pollinators (Johnson et al., 1998; Goldblatt et al., 2001) or the restriction by development constraints in floral development (Armbruster, 2002; Herrera et al., 2002). Recent advances in understanding the molecular mechanisms of floral zygomorphy indicate that the *CYC2*-like genes in the TCP gene family have played an important role to control the floral zygomorphy in flowering plants based on studies of function and expression (Broholm et al., 2008; Busch & Zachgo, 2007; Cubas, Vincent, & Coen, 1999; Feng et al., 2006; Luo et al., 1999; Luo, Carpenter, Vincent, Copsey, & Coen, 1996; Wang et al., 2008).

The *Schizanthus* genus of the Solanaceae family consists of 12 species that are common in Chile and are pollinated by bee-, hummingbird-, and moth (Perez, Arroyo, Medel, & Hershkovitz, 2006). Flowers of *Schizanthus* have a MAB-36-CD process to develop floral zygomorphy, which displays one dorsal petal, two lateral petals, and two ventral petals (Fig. 12). The petals are fused at the base of the corolla forming a tubular shape, which fused to the stamens and staminodes as well. There are five stamens located relatively to the petals; two aborted dorsal, two functional lateral, and one aborted ventral stamen (Fig. 12). Floral morphology of *Schizanthus* is strongly associated with the pollination syndrome (Coocucci, 1989). The bee-pollinated species of *Sc. pinnatus* is an annual with 20 – 50 cm in height. The flower colors of *Sc. Pinnatus* are white, pink, or violate, 2-3 cm in diameter that arrange in panniculate or dichotomous inflorescences (Coocucci, 1989). *Sc. pinnatus* have fringed brightly colored dorsal and lateral petals for attraction, and two fused ventral petals forming a lobed keel that encloses the two functional stamens (Coocucci, 1989) (Fig. 12). During floral development, the fused ventral petals increase in size in coordination with the development of the two functional stamens that form an explosive pollen release mechanism. At maturity, the flower releases the stamens to explosively discharge pollen onto the body of the bee once it lands on the ventral lobed petals. The floral morphology that has given rise to this explosive pollen release occurs uniquely in *Schizanthus* and depends on highly regulated development process of floral zygomorphy (Coocucci, 1989) (Fig. 12 A).

A recent finding indicates that *CYC2*-like genes is not involved in floral zygomrophy development in Solanaceae (J. Zhang & Zhang, 2015). Here, I examine differential gene expression profiles across the zygomorphic corolla of *Sc. pinnatus* using RNA transcriptome sequencing to identify candidate genes responsible for corolla zygomorphy in *Schizanthus* in Solanaeae. In this experiment I ask three questions: (1) Which are the TCP genes expressed in corolla? (2) Are *CYC2* genes expressed in zygomorphic corolla of *S. pinnatus*? and (3) What are the genes up regulated and down regulated in ventral region of zygomorphic corolla of *S. pinnatus*?

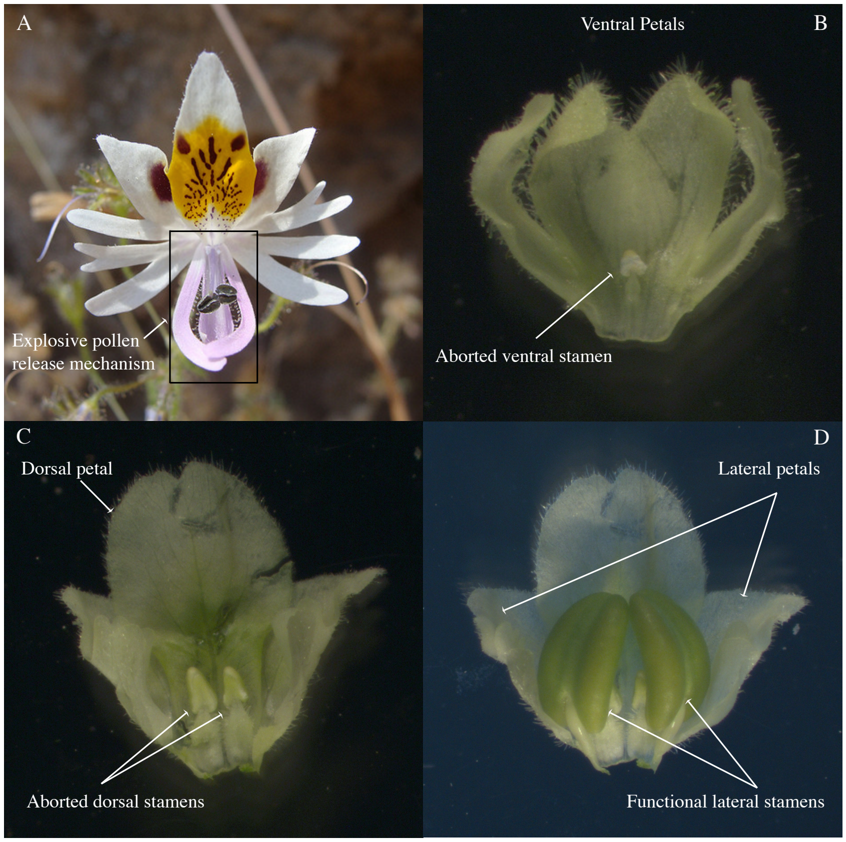


Figure 12. Floral morphology of *Schizanthus pinnatus*. A. Mature flower at late stage of development that has the explosive pollen release mechanism. *Sc. Pinnatus* bud (4 mm in length) that shows in B. the fused ventral petals and the aborted ventral stamen. C. The dorsal petal of the bud and the aborted dorsal stamens. D. The lateral petals and the functional lateral stamens.

Material Method

Plant materials:

Plants of S. pinnatus were grown from seeds at 15°C with 14 hours light and 10 hours darkness cycles in the growth chamber for three months to yield flowers. Plants were watered every three days.

Sampling:

We sampled the young buds of ~4 mm in size for total of 38 buds. The buds were dissected into: dorsal region of corolla (one dorsal petal plus two lateral petals), ventral region of corolla (two ventral petals), dorsal region of androecium (2 dorsal aborted stamens), lateral region of androecium (two functional lateral stamens), and ventral region of androecium (single ventral stamen) (Fig. 12). In current experiment, I used only the dorsal region and ventral region of corolla (Fig. 12). The microdissected samples were frozen in liquid nitrogen and then transfer to -80 for storage.

Total RNA extraction and quality tests:

The dorsal and ventral regions of corolla of micro-dissected samples from the 38 buds were divided into two biological replicates (1.dp1-2, 2.dp2-2, 3.vp1-2, 4.vp2-2). Total RNA was isolated using RNAqueous Total RNA Isolation Kit from Ambion ("Ambion, Austin, TX, USA,"). The quality and the quantity of the isolated 4 total RNA samples were checked using 2% agarose gels, and Nanodrop ("IMPLEN, CA, USA,"), respectively. Then, the total RNA samples were treated with TURBO DNA-free Kit ("Ambion, Austin, TX, USA,") to remove any genomic DNA contamination. The final DNA-free total RNA samples were ≥ 3 μg total, ≥ 80 ng/μL, A260/A280=1.8-2.2, RIN >7.0, prepared in nuclease-free water. These samples were submitted on dry ice to GENEWIZ, LLC ("South Plainfield, NJ, USA,"). After receiving the samples, the quality control (QC) of the RNA samples concentration from *Sc. pinnatus* were further checked by GENEWIZ using three methods: Nanodrop, Qubit RNA Assay, and Bioanalyzer (Table. 6). RNA libraries were multiplexed and subjected to high output mode sequencing on the Illumina HiSeq2500 platform with a 2x100bp paired-end configuration in high output mode (V4 Chemistry), with a total of at least 250 million reads per lane. The *de novo* transcriptome assembly and mapping and hit count measurement were carried out using CLC Genomic Workbench ("CLC Bio-Qiagen ").

Table 6. GENEWIZ Quality Control (QC) report.

Sample Name	GENE-WIZ ID	Vol. (µl)	Nanodrop 2000						Qubit				Bioanalyzer
			Dilut-ion Factor	Nucleic Acid Conc. (ng/µl)	Actual Nucleic Acid Conc. (ng/µl)	Total Amt (ng)	A260/ A280	A260/ A230	Dilution Factor	Nucleic Acid Conc. (ng/µl)	Actual Nucleic Acid Conc. (ng/µl)	Total Amt (ng)	RIN (for RNA)
1. dp1-2	WZ05-1-dp1-2	23	9	71.3	641.7	14759.1	2.07	1.83	9	70.6	635.4	14614.2	7.6
2. dp2-2	WZ06-2-dp2-2	23	9	61.8	556.2	12792.6	2.07	1.83	9	60	540	12420	7.3
3. vp1-2	WZ07-3-vp1-2	19	5	86.1	430.5	8179.5	2.04	1.66	5	79.4	397	7543	7.4
4. vp2-2	WZ08-4-vp2-2	18	4	82.6	330.4	5947.2	2.03	1.56	4	76.2	304.8	5486.4	7.5

Results and discussion

<u>*RNA-seq data analysis:*</u>

S. pinnatus is a non-model species that lacks a reference genome; therefore, *de novo* transcriptome assembly was required for testing gene expression. After trimming low quality bases from sequence data for the pooled four samples, *de novo* transcriptome assembly was conducted using CLC Genomics Workbench 7.0 ("CLC Bio-Qiagen "). The results of *de novo* transcriptome assembly were later used as a reference for mapping and hit count measurement analysis for differential gene expression analysis. The *de novo* transcriptome assembly, mapping, and hit count measurement analysis, and differential gene expression analysis were performed by Genewiz LLC ("South Plainfield, NJ, USA,").

The *de novo* transcriptome assembly resulted 52,149 transcripts. The mean of the length of assembled transcripts was 1,072 base pairs (bp) and the longest transcript was 11,144 bp. The total length of all transcripts was 55.9 Mbp. Consensus sequences for the assembled transcripts were blasted against NCBI nucleotide database by GENEWIZ (Table. 7). FASTq raw data files were delivered from GENEWIZ.

The mapping and hit count measurement analysis was carried out using the assembled 52,149 transcript sequences as a reference for gene expression analysis of the four individual samples. An excel file for each sample (1.dp1-2, 2.dp2-2, 3.vp1-2, 4.vp2-2) of the gene hit counts and reads per kilobase of transcript per million mapped reads (RPKM) values were generated ("South Plainfield, NJ, USA,") (Table. 8).

Table 7. The assembled transcript measurements.

Criterion	Length
N75	762
N50	1,167
N25	1,917
Minimum	455
Maximum	11,144
Average	1,072
Count	52,149
Total	55,902,509

Table 8. The mapping summary.

Sample	Total Fragments	Mapped Fragments	Non-mapped Fragments	% of Mapped Fragments over Total
1dp1-2	69,739,011	37,965,679	31,773,332	54.44
2dp2-2	66,845,729	36,368,881	30,476,848	54.41
3vp1-2	70,893,247	38,280,753	32,612,494	54.00
4vp2-2	64,506,702	34,715,184	29,791,518	53.82

TCP gene expression in corolla:

To determine if members of TCP gene family were differentially expressed across the corolla, the TCP transcripts were identified by two strategies. First, we searched for the TCP genes from GENEWIZ's blast results. Second, we used 26 tomato TCP members as query sequences to blast a local database of all transcripts sequenced from corolla of *Sc. pinnatus* using BLAST+ 2.4.0 ("Camacho et al., 2008,"). A total of 12 unique transcripts belonging to the TCP gene family were detected (Table. 9). Phylogenetic reconstruction including all TCP members from *Solanum lycopersicum* and *Arabidopsis thaliana* placed these 12 transcripts on diverse clades of the TCP gene family (Table. 9). Most interestingly, the RNA-seq results confirmed previous results on low level of *CYC2* expression in corolla using RNA *in situ* hybridization in which no *CYC2* transcripts were detected (Table. 9). Fold changes indicated none of the TCP transcripts significantly differentially expressed across corolla of *Sc. pinnatus* (Table. 9).

Table 9. TCP loci identified from corolla of *Schizanthus pinnatus.*

#	RNA seq transcript ID	Closely related loci from Genebank	Clades of TCP gene family	Fold Change
1	8422	*AtTCP7/21; SlTCP14*	*Class I*	*1.375579869*
2	9424	*AtTCP9/19; SlTCP20*	*Class I*	*1.222765098*
3	4245	*AtTCP9; SlTCP21*	*Class I*	*1.326731176*
4	21557	*AtTCP8/23; SlTCP13*	*Class I*	*2.338903453*
5	21558	*AtTCP8/23; SlTCP13*	*Class I*	*2.746579063*
6	19573	*AtTCP3/4; SlTCP10*	*Class II-CIN (-I)*	*1.013329593*
7	19490	*AtTCP5/13/17; SlTCP4/5/6/28*	*Class II-CIN (-II)*	*-1.794877652*
8	19491	*AtTCP5/13/17; SlTCP4/5/6/28*	*Class II-CIN (-II)*	*-1.612177804*
9	47392	*SlTCP2*	*Class II-CYC/TB1-CYC1*	*-6.553942957*
10	49450	*SlTCP1*	*Class II-CYC/TB1-CYC1*	*1.652948981*
11	31343	*SlTCP3*	*Class II-CYC/TB1-CYC3*	*1.877277771*
12	31344	*SlTCP22*	*Class II-CYC/TB1-CYC3*	*1.289300205*

Notes: (*At*) *Arabidopsis thaliana*; (*Sl*) *Solanum lycopersicum.*

Differential gene expression in corolla:

Gene expression comparison between dp1 and vp1, and between dp2 and vp2 was conducted after quantile normalization. A gene list was selected if the fold-change for normalized RPKM value > 5 for each comparison ("South Plainfield, NJ, USA,"). Since the comparison is between two samples, p-value cannot be calculated, 5 fold-change had been used as a threshold to select genes that have different expression level.

I compared the two excel files of dp1 and vp1, and dp2 and vp2 to summarize the fold changes in the two biological replicates. I wrote codes using Python programming language to compare the fold of changes for sharing transcripts found in both biological samples to determine if the results are consistent between the two samples (Fig. 13). After the comparison, I excluded the not differentially expressed genes and further analyzed the differentially expressed ones. I categorized the differentially expressed genes to; similar genes in both biological replicates in terms of regulation (either both are up-regulated or down-regulated), not similar regulated genes (one up-regulated and one down-regulated), and present/absent genes (invalid number of one gene (absent gene) and a valid number for the other). I excluded the absent/present genes if the valid number is low. I determined which genes are up regulated and down regulated in both biological samples manually with a threshold of > 5 fold-change (Table. 10). To determine the differential gene expression in present/absent transcripts genes, a threshold of > 50 of total genes read was selected (Table. 11).

```
import sys

if len(sys.argv)<4:
   print"USAGE:Python %s filename1 filename2  outputfile" % (sys.argv[0])
   sys.exit

filename1= (sys.argv[1])
filename2= (sys.argv[2])
outputfile= (sys.argv[3])

FNH1= open(filename1)
header= FNH1.readline()
header= header.strip()

FNH2= open(filename2)
header= FNH2.readline()
header= header.strip()

OFH= open(outputfile,'w')
print>>OFH, "(1)Feature ID\t(1)Fold Change\t(1)dp1 total gene reads\t(1)vp1 total
gene reads(2)Feature ID\t(2)Fold Change\t(2)dp2 total gene reads\t(2)vp2 total gene
reads"

TransDict= {}

for line in FNH1:
    line= line.strip()
    #print line
    data= line.split()
    id= data[0]
    num= data[1]
    #print id1
    #print num1
    TransDict[id]=line
    #print TransDict

FNH1.close()


for line in FNH2:
    line= line.strip()
    data= line.split()
    id= data[0]
    num= data[1]
    #print id2
    #print num2
    if id in TransDict:
        TID= TransDict[id]
         print>> OFH,(TID, line)
```

Figure 13. Codes using Python programming language. The code used to compare the fold change of the biological replicates transcripts.

Table 10. Number of up-regulated and down-regulated genes in both biological replicates:

	Consistent gene expression in dp1-vp1&dp2-vp2	
	Down-regulated VP genes	Up-regulated VP genes
Number of Transcripts genes	89	79

Table 11. The present/absent transcripts genes:

	Present-absent genes in dp1-vp1&dp2-vp2			
	present-dp/absent-vp genes		present-vp/absent-dp genes	
	Down-regulated VP genes	Up-regulated VP genes	Down-regulated VP genes	Up-regulated VP genes
Number of Transcripts genes	5	0	1	1

References

Albach, D. C., Meudt, H. M., & Oxelman, B. . (2005). Piecing together the "New" Plantaginaceae. *American Journal of Botany, 92*, 297–315.

Alverson, W. S. , Karol, K. G. , Baum, D. A. , Chase, M. W. , Swensen, S. M. , Mccourt, R., & Sytsma, K. J. . (1998). Circumscription of the Malvales and relationships to other Rosidae: Evidnce from rbcL sequence data. *American Journal of Botany, 85*, 876–887.

. Ambion, Austin, TX, USA. *TURBO DNA-free Kit*

. Ambion, Austin, TX, USA. *RNAqueous Total RNA Isolation Kit*

Applequist, W. L. , & Wallace, R. S. . (2001). Phylogeny of the Portulacaceous cohort based on ndhF sequence data. *Systematic Botany, 26*, 406-419.

Aversano, Riccardo, Contaldi, Felice, Ercolano, Maria Raffaella, Grosso, Valentina, Iorizzo, Massimo, Tatino, Filippo, . . . Ferrarini, Alberto. (2015). The Solanum commersonii genome sequence provides insights into adaptation to stress conditions and genome evolution of wild potato relatives. *The Plant Cell, 27*(4), 954-968.

Backlund, M., Oxelman, B., & Bremer, B. (2000). Phylogenetic relationships within the Gentianales based on ndhF and rbcL sequences, with particular reference to the Loganiaceae. *American Journal of Botany, 87*, 1029-1043.

Bendiksby, M., Thorbek, L., Scheen, A. C., Lindqvist, C., & Ryding, O. (2011). An updated phylogeny and classification of Lamiaceae subfamily Lamioideae. *Taxon, 60*(2), 471-484.

Bombarely, Aureliano, Moser, Michel, Amrad, Avichai, Bao, Manzhu, Bapaume, Laure, Barry, Cornelius S, . . . Bruggmann, Rémy. (2016). Insight into the evolution of the Solanaceae from the parental genomes of Petunia hybrida. *Nature plants, 2*, 16074.

Broholm, S. K., Tahtiharju, S., Laitinen, R. A., Albert, V. A., Teeri, T. H., & Elomaa, P. (2008). A TCP domain transcription factor controls flower type specification along the radial axis of the Gerbera (Asteraceae) inflorescence. *Proc Natl Acad Sci U S A, 105*(26), 9117-9122. doi: 10.1073/pnas.0801359105

Bruneau, A., Doyle, J. J. , Herendeen, P., Hughes, C. , Kenicer, G. , Lewis, G. , . . . Wyk, B. V. . (2013). Legume phylogeny and classification in the 21st century:
Progress, prospects and lessons for other species-rich clades. *Taxon, 62*, 217–248.

Bruneau, A., Klitgaard, B. B., Fougere-Danezan, M., & Tucker, S. C. (2005). *Phylogenetic analysis of floral ontogenetic and molecular characters in Caesalpinioideae: insights into floral evolution in the basal Leguminosae*. Paper presented at the XVII International Botanical Congress, Vienna, Austria.

Bukhari, G., Zhang, J., & Zhang, W. . (2015). *Floral orientation and evolution of flower symmetry in pentapetalous angiosperms* http://2015.botanyconference.org/engine/search/index.php?func=detail&aid=694. Paper presented at the Botany 2015, Edmonton, Alberta - Canada.

Busch, A., Horn, S., & Zachgo, S. (2014). Differential transcriptome analysis reveals insight into monosymmetric corolla development of the crucifer Iberis amara. *Bmc Plant Biology, 14*. doi: Artn 285

10.1186/S12870-014-0285-4
Busch, A., & Zachgo, S. (2007). Control of corolla monosymmetry in the Brassicaceae Iberis amara. *Proc Natl Acad Sci U S A, 104*(42), 16714-16719. doi: 10.1073/pnas.0705338104
. Camacho et al., 2008. *BLAST+ 2.4.0*
Campbell, C. S., Evans, R. C., Morgan, D. R., Dickinson, T. A., & Arsenault, M. P. . (2007). Phylogeny of subtribe Pyrinae (formerly the Maloideae, Rosaceae): Limited resolution of a complex evolutionary history. *Plant Systematics and Evolution, 266*, 119–145.
Citerne, H., Jabbour, F., Nadot, S., & Damerval, C. (2010). The Evolution of Floral Symmetry. *Advances in Botanical Research, Vol 54, 54*, 85-137. doi: 10.1016/S0065-2296(10)54003-5
Clausing, G., & Renner, S. S. (2001). Molecular phylogenetics of Melastomataceae and Memecylaceae: implications for character evolution *American Journal of Botany, 88*, 486–498.
CLC Bio-Qiagen).
Consortium, Potato Genome Sequencing. (2011). Genome sequence and analysis of the tuber crop potato. *Nature, 475*(7355), 189-195.
Consortium, Tomato Genome. (2012). The tomato genome sequence provides insights into fleshy fruit evolution. *Nature, 485*(7400), 635-641.
Coocucci, A. A. (1989). El mecanismo floral de Schizanthus (Solanaceae) (The floral mechanism of Schizanthus Solanaceae). *Kurtziana, 20*, 113-132.
Crane, P. R., Friis, E. M., & Pedersen, K. R. (1995). The Origin and Early Diversification of Angiosperms. *Nature, 374*(6517), 27-33. doi: Doi 10.1038/374027a0
Crepet, W. L. (1996). Timing in the evolution of derived floral characters: Upper Cretaceous (Turonian) taxa with tricolpate and tricolpate-derived pollen. *Review of Palaeobotany and Palynology, 90*(3-4), 339-359. doi: Doi 10.1016/0034-6667(95)00091-7
Cubas, P., Vincent, C., & Coen, E. (1999). An epigenetic mutation responsible for natural variation in floral symmetry. *Nature, 401*(6749), 157-161. doi: 10.1038/43657
Davis, C. C., & Anderson, W. R. (2010). A complete generic phylogeny of Malpighiaceae inferred from nucleotide sequence data and morphology. *American Journal of Botany, 97*(12), 2031-2048. doi: 10.3732/ajb.1000146
De Craene, L. P. R. (2010). *Floral Diagrams*: Cambridge University Press.
De Craene, L. P. R., Soltis, P. S., & Soltis, D. E. (2003). Evolution of floral structures in basal angiosperms. *International Journal of Plant Sciences, 164*(5), S329-S363.
Deng, J. B., Drew, B. T., Mavrodiev, E. V., Gitzendanner, M. A., Soltis, P. S., & Soltis, D. E. (2015). Phylogeny, divergence times, and historical biogeography of the angiosperm family Saxifragaceae. *Mol Phylogenet Evol, 83*, 86-98. doi: 10.1016/j.ympev.2014.11.011
Donoghue, M. J. , Ree, R. H. , & Baum, D. A. . (1998). Phylogeny and the evolution of flower symmetry in the Asteridae. *trends in plant science, 3*, 311–317. doi: 10.1016/S1360-1385(98)01278-3
Eddie, W. M. M., Shulkina, T., Goskin, J., Haberle, R. C., & Jansen, R. K. (2003). Phylogeny of Campanulaceae S. Str. inferred from ITS sequences of nuclear ribosomal DNA. *Annals of the Missouri Botanical Garden, 90*, 554-575.

Eichler, A. W. (1875-78). *Blüthendiagramme construirt und erläutert. [Flowers diagrams constructed and explained] 2 theile in 1*. Leipzig.: Wilhelm Engelmann.
Endress, P. K. (2001). Evolution of floral symmetry. *Current Opinion in Plant Biology, 4*(1), 86-91. doi: Doi 10.1016/S1369-5266(00)00140-0
Engler, A., & Pranrl, K. (1895). *9-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. IV, division 3b)*: Leipzig.
Engler, A., & Pranrl, K. (1897a). *12-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. IV, division 4,5)*: Leipzig.
Engler, A., & Pranrl, K. (1897b). *13-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. IV, division 1,2)*: leipzig.
Engler, A., & Pranrl, K. (1889a). *1-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol.II in 6 division)* Leipzig.
Engler, A., & Pranrl, K. (1889b). *2-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III)*: Leipzig.
Engler, A., & Pranrl, K. (1889c). *3-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 1)*: Leipzig.
Engler, A., & Pranrl, K. (1891a). *4-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 2)*: Leipzig.
Engler, A., & Pranrl, K. (1891b). *5-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 2a)*: Leipzig.
Engler, A., & Pranrl, K. (1893). *6-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 1a)*: Leipzig.
Engler, A., & Pranrl, K. (1894). *7-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 3)*: Leipzig.

Engler, A., & Pranrl, K. (1895). *8-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 6,6a)*: Leipzig.
Engler, A., & Pranrl, K. (1897a). *10-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 4,5)*: Leipzig.
Engler, A., & Pranrl, K. (1897b). *11-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. IV, division 3a)*: Leipzig.
Engler, A., & Pranrl, K. (1898). *14-Die natürlichen Pflanzen Familien nebst ihren Gattungen und wichtigeren Arten insbesondere den Nutzpflanzen (The natural plant families in addition to their genera and species important in particular to crops) (Vol. III, division 7,8)*: Leipzig.
Feng, X., Zhao, Z., Tian, Z., Xu, S., Luo, Y., Cai, Z., . . . Luo, D. (2006). Control of petal shape and floral zygomorphy in Lotus japonicus. *Proc Natl Acad Sci U S A, 103*(13), 4970-4975. doi: 10.1073/pnas.0600681103
Fior, S., Karis, P. O., Casazza, G., Minuto, L., & Sala, F. (2006). Molecular phylogeny of the Caryophyllaceae (Caryophyllales) inferred from chloroplast matK and nuclear rDNA ITS sequences. *American Journal of Botany, 93*(3), 399-411. doi: 10.3732/ajb.93.3.399
Giurfa, M., Dafni, A., & Neal, P. R. (1999). Floral Symmetry and Its Role in Plant-Pollinator Systems. *International Journal of Plant Sciences, 160*(S6), S41-S50.
Goetsch, L., Eckert, A. J. , & Hall, B. D. . (2005). The molecular systematics of Rhododendron (Ericaceae): A phylogeny based upon RPB2 gene sequences. *Systematic Botany, 30*, 616-626. doi: http://dx.doi.org/10.1600/0363644054782170
Gomez, J. M., Perfectti, F., & Camacho, J. P. M. (2006). Natural selection on Erysimum mediohispanicum flower shape: Insights into the evolution of zygomorphy. *American Naturalist, 168*(4), 531-545. doi: Doi 10.1086/507048
Gomez, J. M., Perfectti, F., & Lorite, J. (2015). The role of pollinators in floral diversification in a clade of generalist flowers. *Evolution, 69*(4), 863-878. doi: 10.1111/evo.12632
Groppo, M., Pirani, J. R., Salatino, M. L., Blanco, S. R., & Kallunki, J. A. (2008). Phylogeny of Rutaceae based on twononcoding regions from cpDNA. *American Journal of Botany, 95*(8), 985-1005. doi: 10.3732/ajb.2007313
Hileman, L. C. (2014a). Bilateral flower symmetry - how, when and why? *Current Opinion in Plant Biology, 17*, 146-152. doi: 10.1016/j.pbi.2013.12.002
Hileman, L. C. (2014b). Trends in flower symmetry evolution revealed through phylogenetic and developmental genetic advances. *Philosophical Transactions of the Royal Society B-Biological Sciences, 369*(1648). doi: Artn 20130348
10.1098/Rstb.2013.0348

Hoot, S. B. (1991). Phylogeny of the Ranunculaceae based on epidermal microcharacters and macromorphology. *American Society of Plant Taxonomists, 16*, 741-755. doi: 10.2307/2418876

Horn, James W. (2009). Phylogenetics of Dilleniaceae using sequence data from four plastid loci (rbcL,infA,rps4,rpl16Intron). *International Journal of Plant Sciences, 170*(6), 794-813. doi: 10.1086/599239

Hufford, L., Mcmahon, M. M. , Sherwood, A. M. , Reeves, G., & Chase, M. W. . (2003). The major clades of Loasaceae: Phylogenetic analysis using the plastid matK and trnL-trnF regions. *American Journal of Botany, 90*, 1215–1228.

Hufford, L., Moody, M. L., & Soltis, D. E. (2001). A phylogenetic analysis of hydrangeaceae based on sequences of the plastid gene matk and their combination with rbcl and morphological data. *International Journal of Plant Sciences, 162*(4), 835-846. doi: Doi 10.1086/320789

. IMPLEN, CA, USA. *Nanodrop*.

The International Plant Names Index (2012).

Janssens, S. , Geuten, K., Yuan, Y. , Song, Y., Kupfer, P., & Smets, E. (2006). Phylogenetics of Impatiens and Hydrocera (Balsaminaceae) using chloroplast atpB-rbcL spacer sequences. *Systematic Botany, 31*, 171-180. doi: http://dx.doi.org/10.1600/036364406775971796

Kallersjo, M., Bergqvist, G., & Anderberg, A. A. . (2000). Generic realignment in primuloid families of the Ericales s.l.: A phylogenetic analysis basef on DNA sequences from three chloroplast genes and morphology. *American Journal of Botany, 87*, 1325–1341.

Kron, K. A. . (1996). Phylogenetic relationships of Empetraceae, Epacridaceae, Ericaceae, Monotropaceae, and Pyrolaceae: Evidence from nuclear ribosomal 18s sequence data. *Annals of Botany, 77*, 293-303.

Linder, H. P., Dlamini, T., Henning, J., & Verboom, G. A. . (2006). The evolutionary history of melianthus (Melianthaceae) *American Journal of Botany, 93*, 1052–1064.

Lledo, M. D., Crespo, M. B., Fay, M. F., & Chase, M. W. (2005). Molecular phylogenetics of Limonium and related genera (Plumbaginaceae): biogeographical and systematic implications. *American Journal of Botany, 92*(7), 1189-1198. doi: 10.3732/ajb.92.7.1189

Luna, I., & Ochoterena, H. (2004). Phylogenetic relationships of the genera of Theaceae based on morphology. *Cladistics-the International Journal of the Willi Hennig Society, 20*(3), 223-270. doi: Doi 10.1111/J.1096-0031.2004.00024.X

Luo, D., Carpenter, R., Copsey, L., Vincent, C., Clark, J., & Coen, E. (1999). Control of organ asymmetry in flowers of Antirrhinum. *Cell, 99*(4), 367-376.

Luo, D., Carpenter, R., Vincent, C., Copsey, L., & Coen, E. (1996). Origin of floral asymmetry in Antirrhinum. *Nature, 383*(6603), 794-799. doi: 10.1038/383794a0

Maddison, W. P., & Maddison, D.R. . (2015). Mesquite: a modular system for evolutionary analysis. Version 3.03 http://mesquiteproject.org.

Martin-Bravo, S., Meimberg, H., Luceno, M., Markl, W., Valcarcel, V., Brauchler, C., . . . Heubl, G. . (2007). Molecular systematics and biogeography of Resedaceae based on ITS and trnL-F sequences. *Mol Phylogenet Evol, 44*, 1105–1120.

Maurin, O., Chase, M. W., Jordaan, M., & Van Der Bank, M. . (2010). Phylogenetic relationships of Combretaceae inferred from nuclear and plastid DNA sequence data:

implications for generic classification. *Botanical Journal of the Linnean Society, 162*, 453–476.
McDade, L. A., Masta, S. E., Moody, M. L., & Waters, E. (2000). Phylogenetic relationships among Acanthaceae: Evidence from two genomes. *Systematic Botany, 25*(1), 106-121. doi: Doi 10.2307/2666677
McDade, L. A., & Moody, M. L. (1999). Phylogenetic relationships among Acanthaceae: Evidence from noncoding trnL-trnF chloroplast DNA sequences. *American Journal of Botany, 86*(1), 70-80. doi: Doi 10.2307/2656956
Mcdill, J. R., & Simpson, B. B. . (2010). Molecular phylogenetics of Linaceae with complete generic sampling and data from two plastid genes. *Botanical Journal of the Linnean Society, 165*, 64–83.
Moody, M. L. , Hufford, L. , Soltis, D. E. , & Soltis, P. S. . (2001). Phylogenetic relationships of Loasaceae subfamily Gronovioideae inferred from matK and ITS sequence data. *American Journal of Botany, 88*, 326–336.
Moorea, M. J., Soltisb, P. S., Bellc, C. D., Burleighd, J. G., & Soltis, D. E. . (2010). Phylogenetic analysis of 83 plastid genes further resolves the early diversification of eudicots. *PNAS, 107*, 4623–4628.
Morton, C. M. , & Kallunki, J. A. . (1993). Pollen morphology of the subtribe Cuspariinae (Rutaceae) *Brittonia, 45*, 286-314.
Nazaire, M., & Hufford, L. (2012). A Broad phylogenetic analysis of Boraginaceae: Implications for the relationships of Mertensia. *Systematic Botany, 37*(3), 758-783. doi: Doi 10.1600/036364412x648715
Nepokroeff, Molly, & Zimmer, E. A. (2002). "Angiosperms" Biology. Retrieved from Encyclopedia.com website: doi:http://www.encyclopedia.com/doc/1G2-3400700026.html
Neyland, R. . (2001). A phylogeny inferred from large ribosomal subunit (26S) rDNA sequences suggests that Cuscuta is a derived member of Convolvulaceae *Brittonia, 53*, 108-115.
Olmstead, R. G., Bohs, L., Abdel Migid, H., Santiago-Valentin, E., Garcia, V. F., & Collier, S. M. . (2008). A molecular phylogeny of the Solanaceae. *Taxon, 57*, 1159–1181.
Oxelman, B., Kornhall, P., Olmstead, R. G., & Bremer, B. (2005). Further disintegration of Scrophulariaceae *Taxon, 54*, 411-425.
Park, S., & Kim, K. . (2004). Molecular phylogeny of the genus Hypericum (Hypericaceae) from Korea and Japan: Evidence from nuclear rDNA ITS sequence data. *Journal of Plant Biology, 47*, 366-374.
Perez, F., Arroyo, M. T. K., Medel, R., & Hershkovitz, M. A. (2006). Ancestral reconstruction of flower morphology and pollination systems in Schizanthus (Solanaceae). *American Journal of Botany, 93*(7), 1029-1038. doi: Doi 10.3732/Ajb.93.7.1029
The Plant List. (2010).
Potgieter, K., & Albert, V. A. . (2001). Phylogenetic relationships within Apocynaceae s.l. based on trnL intron and trnL-F spacer sequences and propagule characters. *Annals of the Missouri Botanical Garden, 88*, 523-549.

Potter, D., Eriksson, T., Evans, R. C., Oh, S., Smedmark, J. E. E., Morgan, D. R., . . . Campbell, C. S. . (2007). Phylogeny and classification of Rosaceae. *Plant Systematics and Evolution, 266*, 5–43.

Preston, J. C., Martinez, C. C., & Hileman, L. C. (2011). Gradual disintegration of the floral symmetry gene network is implicated in the evolution of a wind-pollination syndrome. *Proc Natl Acad Sci U S A, 108*(6), 2343-2348. doi: 10.1073/pnas.1011361108

Price, R. A., & Palmer, J. D. (1993). Phylogenetic relationships of the Geraniaceae and Geraniales from rbcL sequence comparisons. *Annals of the Missouri Botanical Garden, 80*, 661-671.

Qin, Cheng, Yu, Changshui, Shen, Yaou, Fang, Xiaodong, Chen, Lang, Min, Jiumeng, . . . Luo, Yong. (2014). Whole-genome sequencing of cultivated and wild peppers provides insights into Capsicum domestication and specialization. *Proceedings of the National Academy of Sciences, 111*(14), 5135-5140.

Reyes, E., Nadot, S., & Sauquet, H. (2015). *Floral symmetry shifted 198 times in angiosperms* http://2015.botanyconference.org/engine/search/index.php?func=detail&aid=297. Paper presented at the Botany 2015, Edmonton, Alberta - Canada.

Rivadavia, F., Kondo, K., Kato, M., & Hasebe, M. (2003). Phylogeny of the sundews, Drosera (Droseraceae), based on chloroplast rbcL and nuclear 18S ribosomal DNA Sequences. *American Journal of Botany, 90*(1), 123-130. doi: 10.3732/ajb.90.1.123

Rosin, F. M., & Kramer, E. M. (2009). Old dogs, new tricks: Regulatory evolution in conserved genetic modules leads to novel morphologies in plants. *Developmental Biology, 332*(1), 25-35. doi: 10.1016/j.ydbio.2009.05.542

Ruhfel, B. R., Stevens, P. F., & Davis, C. C. . (2013). Combined morphological and molecular phylogeny of the Clusioid clade (Malpighiales) and the placement of the ancient rosid macrofossil paleoclusia *International Journal of Plant Sciences, 174*, 910–936.

Sayers, E. W., Barrett, T., Benson, D. A., Bryant, S. H., Canese, K., Chetvernin, V., . . . Ye, J. (2009). Database resources of the National Center for Biotechnology Information. *Nucleic Acids Research, 37*, D5-D15. doi: 10.1093/nar/gkn741

Schneider, J. V., Bissiengou, P., Amaral Mdo, C., Tahir, A., Fay, M. F., Thines, M., . . . Chatrou, L. W. (2014). Phylogenetics, ancestral state reconstruction, and a new infrafamilial classification of the pantropical Ochnaceae (Medusagynaceae, Ochnaceae s.str., Quiinaceae) based on five DNA regions. *Mol Phylogenet Evol, 78*, 199-214. doi: 10.1016/j.ympev.2014.05.018

Schonenberger, J., Anderberg, A. A., & Sytsma, K. J. . (2005). Molecular Phylogenetics and patterns of floral evolution in the Ericales. *International Journal of Plant Sciences, 166*, 265-288.

Schultheis, L. M. , & Donoghue, M. J. . (2004). Molecular phylogeny and biogeography of Ribes (Grossulariaceae), with an emphasis on Gooseberries (Subg. Grossularia). *Systematic Botany, 29*, 77-96.

Sheahan, M. C., & Chase, M. W. (2000). Phylogenetic relationships within zygophyllaceae based on DNA sequences of three plastid regions, with special emphasis on zygophylloideae. *Systematic Botany, 25*(2), 371-384. doi: Doi 10.2307/2666648

Simmons, M. P., Savolainen, V., Clevinger, C. C., Archer, R. H., & Davis, J. I. (2001). Phylogeny of the Celastraceae inferred from 26S nuclear ribosomal DNA, phytochrome B, rbcL, atpB, and morphology. *Mol Phylogenet Evol, 19*(3), 353-366. doi: 10.1006/mpev.2001.0937

Sinou, C., Forest, F., Lewis, G. P. , & Bruneaua, A. (2009). The genus Bauhinia s.l. (Leguminosae): a phylogeny based on the plastid trnL–trnF region. *Botany, 87*, 947-960.

Soltis, D. E., Senters, A. E., Zanis, M. J., Kim, S., Thompson, J. D., Soltis, P. S., . . . Farris, J. S. (2003). Gunnerales are sister to other core eudicots: Implications for the evolution of pentamery. *American Journal of Botany, 90*(3), 461-470. doi: Doi 10.3732/Ajb.90.3.461

Soltis, P. S., & Soltis, D. E. (2004). The origin and deversification of angiosperms. *American Journal of Botany, 91*(10), 1614-1626. doi: Doi 10.3732/Ajb.91.10.1614

. South Plainfield, NJ, USA. *GENEWIZ LLC*

Stevens, P. F. . (2001 onwards). Angiosperm Phylogeny Website. Version 12, July 2012 [and more or less continuously updated since]." will do.

Ushimaru, A., Dohzono, I., Takami, Y., & Hyodo, F. (2009). Flower orientation enhances pollen transfer in bilaterally symmetrical flowers. *Oecologia, 160*(4), 667-674. doi: 10.1007/s00442-009-1334-9

Wang, Z., Luo, Y., Li, X., Wang, L., Xu, S., Yang, J., . . . Luo, D. (2008). Genetic control of floral zygomorphy in pea (Pisum sativum L.). *Proc Natl Acad Sci U S A, 105*(30), 10414-10419. doi: 10.1073/pnas.0803291105

Wannan, B.S. (2006). Analysis of generic relationships in Anacardiaceae. *Blumea, 51*, 165–195.

Webb, C. O. , & Donoghue, M. J. (2004). Phylomatic: tree assembly for applied phylogenetics. 5: 181-183 (http://phylodiversity.net/phylomatic/)

Wen, J., Plunkett, G. M., Mitchell, A. D., & Wagstaff, S. J. . (2001). The evolution of Araliaceae: A phylogenetic analysis based on ITS sequences of nuclear ribosomal DNA. *Systematic Botany, 26*, 144-167.

Wilson, P. G. , Heslewood, M. , Lam, N., & Quinn, C. . (2004). Progress towards a phylogeny of the Chamekaucium alliance (Myrtaceae). *Australian Biology*.

Xi, Z., Ruhfel, B. R., Schaefer, H., Amorim, A. M., Sugumaran, M., Wurdack, K. J., . . . Davis, C. C. (2012). Phylogenomics and a posteriori data partitioning resolve the Cretaceous angiosperm radiation Malpighiales. *Proceedings of the National Academy of Sciences, 109*(43), 17519-17524. doi: 10.1073/pnas.1205818109

Yakandawala, D., Morton, C. M., & Prance, G. T. . (2010). Phyogenetic relationships of the Chrysobalanaceae inferred from chloroplast, nuclear, and morphological data. *Annals of the Missouri Botanical Garden, 97*, 259-281.

Zhang, J., & Zhang, W. (2015). *Evolution, expression, and function of CYC2-like genes in Solanaceae.* Paper presented at the Botany 2015, Edmonton, Alberta - Canada.

Zhang, W. H., Chen, Z. D., Li, J. H., Chen, H. B., & Tang, Y. C. (2003). Phylogeny of the Dipsacales s.l. based on chloroplast trnL-F and ndhF sequences. *Mol Phylogenet Evol, 26*(2), 176-189.

APPENDIX

Appendix table 1. Floral characters describe the developmental mechanisms of floral symmetry. Authors are 1: (De Craene, 2010), 2: (A. Engler & K Pranrl, 1895; Engler & Pranrl, 1897a, 1897b; Engler & Pranrl, 1889a, 1889b, 1889c, 1891a, 1891b, 1893, 1894; A. Engler & K. Pranrl, 1895; Engler & Pranrl, 1897a, 1897b, 1898), 3: (Eichler, 1875-78).

Order	Family	Species	Original species	Floral Initiation	Mechanisms	Pattern Symm.	Floral Symm.	Author(s)
Malvales	Malvaceae	*Abutilon megapotamicum*	*Abutilon megapotamicum*	MAB	MAB-A	R	Actino	1
Apiales	Apiaceae	*Aegopodium podagraria*	*Aegopodium podagraria*	MAB	MAB-A	R	Actino	1
Berberidopsidales	Aextoxicaceae	*Aextoxicon punctatum*	*Aextoxicon punctatum*	MAD	MAD-A	R	Actino	1
Fabales	Fabaceae	*Afzelia quanzensis*	*Afzelia quanzensis*	MAD	MAD-0-CD	CD	Zygo	1
Ericales	Primulaceae	*Anagallis arvensis*	*Anagalis arvensis*	MAB	MAB-A	R	Actino	1
Ranunculales	Ranunculaceae	*Aquilegia sp*	*Aquilegia sp*	MAD	MAD-A	R	Actino	1
Oxalidales	Oxalidaceae	*Averrhoa carambola*	*Averrhoa carambola*	MAB	MAB-A	R	Actino	1
Celastrales	Celastraceae	*Brexia madagascariensis*	*Brexia madagascariensis*	MAB	MAB-A	R	Actino	1
Cornales	Loasaceae	*Caiophora hibiscifolia*	*Cajophora hibiscifolia*	MAB	MAB-A	R	Actino	1
Fabales	Fabaceae	*Calliandra haematocephala*	*Calliandra haematocephala*	MAB	MAB-A	R	Actino	1
Myrtales	Myrtaceae	*Callistemon citrinus*	*Callistemon citrinus*	MAB	MAB-A	R	Actino	1
Rosales	Rhamnaceae	*Ceanothus dentatus*	*Ceanothus dentatus*	MAB	MAB-A	R	Actino	1
Caryophyllales	Plumbaginaceae	*Ceratostigma minus*	*Ceratostigma minus*	MAB	MAB-A	R	Actino	1
Solanales	Solanaceae	*Cestrum parqui*	*Cestrum parqui*	MAB	MAB-A	R	Actino	1
Malpighiales	Chrysobalanaceae	*Chrysobalanus sp*	*Chrysobalanus sp*	MAB	MAB-A	R	Actino	1
Malvales	Cistaceae	*Cistus salviifolius*	*Cistus salviifolius*	MAD	MAD-A	R	Actino	1

Lamiales	Lamiaceae	*Clerodendrum petasites*	*Clerodendron petasites*	MAB	MAB-0-CV	CV	Zygo	1
Ericales	Pentaphylacaceae	*Cleyera japonica*	*Cleyera japonica*	MAB	MAB-A	R	Actino	1
Santalales	Olacaceae	*Coula edulis*	*Coula edulis*	MAD	MAD-A	R	Actino	1
Oxalidales	Elaeocarpaceae	*Crinodendron patagua*	*Crinodendron patagua*	MAB	MAB-A	R	Actino	1
Cucurbitales	Cucurbitaceae	*Cucurbita palmata*	*Cucurbita palmata*	MAB	MAB-A	R	Actino	1
Lamiales	Scrophulariaceae	*Diascia vigilis*	*Diascia vigilis*	MAB	MAB-0-CV	CV	Zygo	1
Cornales	Hydrangeaceae	*Dichroa febrifuga*	*Dichroa febrifuga*	MAB	MAB-A	R	Actino	1
Caryophyllales	Droseraceae	*Drosera capensis*	*Drosera capensis*	MAD	MAD-A	R	Actino	1
Boraginales	Boraginaceae	*Echium hierrense*	*Echium hierrense*	MAB	MAB-144-CV	CV	Zygo	1
Geraniales	Geraniaceae	*Erodium leucanthum*	*Erodium leucanthemum*	MAB	MAB-0-CV	CV	Zygo	1
Gentianales	Gentianaceae	*Exacum affine*	*Exacum affine*	MAB	MAB-108-CD	CD	Zygo	1
Caryophyllales	Frankeniaceae	*Frankenia laevis*	*Frankenia laevis*	MAB	MAB-A	R	Actino	1
Sapindales	Rutaceae	*Galipea riedeliana*	*Galipea riedeliana*	MAB	MAB-A	R	Actino	1
Malpighiales	Malpighiaceae	*Galphimia glauca*	*Galphimia glauca*	MAB	MAB-36-CD	CD	Zygo	1
Myrtales	Combretaceae	*Guiera senegalensis*	*Guiera senegalensis*	MAB	MAB-A	R	Actino	1
Dilleniales	Dilleniaceae	*Hibbertia cuneiformis*	*Hibbertia cuneiformis*	MAB	MAB-A	R	Actino	1
Malpighiales	Hypericaceae	*Hypericum perforatum*	*Hypericum perforatum*	MAB	MAB-A	R	Actino	1
Ericales	Balsaminaceae	*Impatiens platypetala*	*Impatiens platypetala*	MAB	MAB-180-CD	CD	Zygo	1
Ericales	Primulaceae	*Jacquinia macrocarpa*	*Jacquinia macrocarpa*	MAB	MAB-A	R	Actino	1
Malpighiales	Euphorbiaceae	*Jatropha sp*	*Jatropha sp*	MAB	MAB-A	R	Actino	1
Cornales	Hydrangeaceae	*Kirengeshoma palmata*	*Kirengeshoma palmata*	MAD	MAD-A	R	Actino	1
Malpighiales	Phyllanthaceae	*Leptopus chinensis*	*Leptopus chinensis*	MAB	MAB-A	R	Actino	1
Asterales	Asteraceae	*Ligularia dentata*	*Ligularia dentata*	MAB	MAB-A	R	Actino	1
Malpighiales	Linaceae	*Linum monogynum*	*Linum monogynum*	MAB	MAB-A	R	Actino	1
Asterales	Campanulaceae	*Lobelia tupa*	*Lobelia tupa*	MAD	MAD-180-CV	CV	Zygo	1
Dipsacales	Caprifoliaceae	*Lonicera giraldii*	*Lonicera giraldii*	MAB	MAB-0-CV	CV	Zygo	1
Ericales	Ericaceae	*Macleania stricta*	*Macleania stricta*	MAB	MAB-A	R	Actino	1
Myrtales	Melastomataceae	*Medinilla magnifica*	*Medinilla magnifica*	MAB	MAB-A	R	Actino	1
Geraniales	Melianthaceae	*Melianthus major*	*Melianthus major*	MAB	MAB-180-CD	CD	Zygo	1
Sabiales	Sabiaceae	*Meliosma dilleniifolia*	*Meliosma dilleniifolia*	MAD	MAD-72-CD	CD	Zygo	1
Brassicales	Moringaceae	*Moringa oleifera*	*Moringa oleifera*	MAB	MAB-108-CD	CD	Zygo	1
Ericales	Lecythidaceae	*Napoleonaea vogelii*	*Napoleonaea vogelii*	MAB	MAB-A	R	Actino	1
Gentianales	Apocynaceae	*Nerium oleander*	*Nerium oleander*	MAB	MAB-A	R	Actino	1
Malvales	Neuradaceae	*Neurada procumbens*	*Neurada procumbens*	MAD	MAD-A	R	Actino	1
Malpighiales	Ochnaceae	*Ochna multiflora*	*Ochna multiflora*	MAB	MAB-A	R	Actino	1

Lamiales	Acanthaceae	*Odontonema strictum*	*Odontonema strictum*	MAB	MAB-0-CV	CV	Zygo	1
Liliales	Melanthiaceae	*Paris polyphylla*	*Paris polyphylla*	MAB	MAB-A	R	Actino	1
Malpighiales	Passifloraceae	*Passiflora vitifolia*	*Passiflora vitifolia*	MAB	MAB-A	R	Actino	1
Geraniales	Geraniaceae	*Pelargonium abrotanifolium*	*Pelargonium abrotanifolium*	MAB	MAB-0-CV	CV	Zygo	1
Lamiales	Plantaginaceae	*Penstemon fruticosus*	*Penstemon fruticosus*	MAB	MAB-0-CV	CV	Zygo	1
Gentianales	Apocynaceae	*Periploca graeca*	*Periploca graeca*	MAB	MAB-A	R	Actino	1
Apiales	pittosporaceae	*Pittosporum tobira*	*Pittosporum tobira*	MAB	MAB-A	R	Actino	1
Fabales	Polygalaceae	*Polygala myrtifolia*	*Polygala xdalmaisiana*	MAB	MAB-0-CV	CV	Zygo	1
Caryophyllales	Tamaricaceae	*Reaumuria vermiculata*	*Reaumuria vermiculata*	MAB	MAB-A	R	Actino	1
Saxifragales	Grossulariaceae	*Ribes speciosum*	*Ribes speciosum*	MAB	MAB-A	R	Actino	1
Saxifragales	Saxifragaceae	*Saxifraga fortunei*	*Saxifraga fortunei*	MAB	MAB-36-CD	CD	Zygo	1
Apiales	Araliaceae	*Schefflera elliptica*	*Scheffleria elliptica*	MAB	MAB-A	R	Actino	1
Sapindales	Sapindaceae	*Serjania glabrata*	*Serjania glabrata*	MAB	MAB-72-CV	CV	Zygo	1
Ericales	Primulaceae	*Soldanella villosa*	*Soldanella villosa*	MAB	MAB-A	R	Actino	1
Rosales	Rosaceae	*Spiraea salicifolia*	*Spiraea salicifolia*	MAB	MAB-A	R	Actino	1
Ericales	Theaceae	*Stewartia pseudocamellia*	*Stewartia pseudocamellia*	MAB	MAB-A	R	Actino	1
Fabales	Fabaceae	*Strongylodon macrobotrys*	*Strongylodon macrobotrys*	MAD	MAD-0-CD	CD	Zygo	1
Asterales	Stylidiaceae	*Stylidium graminifolium*	*Stylidium graminifolium*	MAB	MAB-0-CV	CV	Zygo	1
Malpighiales	Clusiaceae	*Symphonia globulifera*	*Symphonia globulifera*	MAB	MAB-A	R	Actino	1
Malvales	Malvaceae	*Theobroma cacao*	*Theobroma cacao*	MAB	MAB-A	R	Actino	1
Santalales	Santalaceae	*Thesium strictum*	*Thesium strictum*	MAB	MAB-A	R	Actino	1
Malvales	Malvaceae	*Tilia europaea*	*Tilia europaea*	MAB	MAB-A	R	Actino	1
Celastrales	Celastraceae	*Tontelea sp*	*Tonteleia sp*	MAB	MAB-A	R	Actino	1
Sapindales	Anacardiaceae	*Toxicodendron vernicifluum*	*Toxicodendron vernicifluum*	MAB	MAB-A	R	Actino	1
Dipsacales	Caprifoliaceae	*Valeriana repens*	*Valeriana repens*	MAB	MAB-0-CV	CV	Zygo	1
Dipsacales	Adoxaceae	*Viburnum grandiflorum*	*Viburnum grandiflorum*	MAB	MAB-A	R	Actino	1
Malpighiales	Violaceae	*Viola tricolor*	*Viola tricolor*	MAB	MAB-0-CV	CV	Zygo	1
Malpighiales	Phyllanthaceae	*Andrachne telephioides*	*Andrachne telephioides*	MAB	MAB-A	R	Actino	2
Gentianales	Loganiaceae	*Antonia ovata*	*Antonia ovata*	MAB	MAB-A	R	Actino	2
Caryophyllales	Plumbaginaceae	*Armeria plantaginea*	*Armeria plantaginea*	MAB	MAB-A	R	Actino	2
Malpighiales	Malpighiaceae	*Aspidopterys roxburghiana*	*Aspidopteris roxburghiana*	MAB	MAB-A	R	Actino	2
Saxifragales	Saxifragaceae	*Astilbe thunbergii*	*Astilbe thunbergii*	MAD	MAD-A	R	Actino	2
Malvales	Malvaceae	*Ayenia cordobensis*	*Ayenia cordobensia*	MAB	MAB-A	R	Actino	2
Fabales	Fabaceae	*Bauhinia forficata*	*Bauhinia forficata*	MAD	MAD-A	R	Actino	2
Fabales	Fabaceae	*Bauhinia scandens*	*Bauhinia anguinea*	MAD	MAD-A	R	Actino	2

Fabales	Fabaceae	*Bauhinia monandra*	*Bauhinia krugii*	MAD	MAD-A	R	Actino	2
Fabales	Fabaceae	*Bauhinia pauletia*	*Bauhinia pauletia*	MAD	MAD-A	R	Actino	2
Fabales	Fabaceae	*Bauhinia purpurea*	*Bauhinia purpurea*	MAD	MAD-A	R	Actino	2
Fabales	Fabaceae	*Bauhinia tomentosa*	*Bauhinia tomentosa*	MAD	MAD-A	R	Actino	2
Geraniales	Melianthaceae	*Bersama maxima*	*Bersama maxima*	MAB	MAB-0-CV	CV	Zygo	2
Bruniales	Bruniaceae	*Berzelia lanuginosa*	*Berzelia lanuginosa*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Blastemanthus gemmiflorus*	*Blastemanthus gemmiflorus*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Blumenbachia hieronymi*	*Blumenbachia hieronymi*	MAB	MAB-A	R	Actino	2
Bruniales	Bruniaceae	*Brunia nodiflora*	*Brunia nodiflora*	MAB	MAB-A	R	Actino	2
Caryophyllales	Portulacaceae	*Calandrinia ciliata*	*Calandrinia menziesii*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Callistemon lanceolatus*	*Callistemon lanceolatus*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Calytrix tetragona*	*Calycothrix tetragona*	MAB	MAB-A	R	Actino	2
Malpighiales	Malpighiaceae	*Camarea hirsuta*	*Camarea hirsuta*	MAB	MAB-36-CD	CD	Zygo	2
Na	Fabaceae	*Cassia bacillaris*	*Cassia bacillaris*	MAD	MAD-A	R	Actino	2
Malvales	Malvaceae	*Ceiba pentandra*	*Ceiba risieri*	MAD	MAD-A	R	Actino	2
Celastrales	Celastraceae	*Celastrus paniculatus*	*Celastrus panniculata*	MAB	MAB-A	R	Actino	2
Caryophyllales	Plumbaginaceae	*Ceratostigma plumbaginoides*	*Ceratostigma plumbaginoides*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Chamelaucium ciliatum*	*Chamaelaucium cilitium*	MAB	MAB-A	R	Actino	2
Ericales	Cyrillaceae	*Cliftonia ligustrina*	*Cliftonia ligustrina*	MAB	MAB-A	R	Actino	2
Caryophyllales	Molluginaceae	*Corrigiola litoralis*	*Corriola litoralis*	MAB	MAB-A	R	Actino	2
Oxalidales	Cunoniaceae	*Cunonia capensis*	*Cunonia capensis*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Darwinia oederoides*	*Daruinia oederoides*	MAB	MAB-A	R	Actino	2
Dilleniales	Dilleniaceae	*Davilla rugosa*	*Davilla rugosa*	MAB	MAB-A	R	Actino	2
Ericales	Diapensiaceae	*Diapensia lapponica*	*Diapensia lapponica*	MAB	MAB-A	R	Actino	2
Fabales	Polygalaceae	*Diclidanthera laurifolia*	*Diclidanthera laurifolia*	MAB	MAB-A	R	Actino	2
Icacinales	Icacinaceae	*Emmotum nitens*	*Emmotum nitens*	MAD	MAD-A	R	Actino	2
Ericales	Ericaceae	*Epacris sp*	*Epacris sp*	MAB	MAB-A	R	Actino	2
Malpighiales	Erythroxylaceae	*Erythroxylum sp*	*Erythroxylon paraense*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Eucnide bartonioides*	*Eucnide bartonioides*	MAB	MAB-A	R	Actino	2
Sapindales	Rutaceae	*Zanthoxylum caribaeae*	*Fagara caribaea*	MAB	MAB-A	R	Actino	2
Sapindales	Rutaceae	*Zanthoxylum flava*	*Fagara flava*	MAB	MAB-A	R	Actino	2
Sapindales	Rutaceae	*Zanthoxylum martinicensis*	*Fagara martinicensis*	MAB	MAB-A	R	Actino	2
Sapindales	Rutaceae	*Zanthoxylum monophylla*	*Fagara monophylla*	MAB	MAB-A	R	Actino	2
Malpighiales	Clusiaceae	*Garcinia vilersiana*	*Garcinia vilersiana*	MAD	MAD-A	R	Actino	2
Lamiales	Lamiaceae	*Callicarpa farinosa*	*Geunsia farinosa*	MAB	MAB-A	R	Actino	2

Malpighiales	Ochnaceae	*Godoya antioquiensis*	*Godoya antioquiensis*	MAB	MAB-A	R	Actino	2
Geraniales	Melianthaceae	*Greyia sutherlandii*	*Greyia sutherlandii*	MAB	MAB-A	R	Actino	2
Lamiales	Scrophulariaceae	*Hebenstretia dentata*	*Hehenstreitia dentata*	MAB	MAB-0-CV	CV	Zygo	2
Dilleniales	Dilleniaceae	*Hibbertia hypericoides*	*Hibbertia hyperieoides*	MAB	MAB-A	R	Actino	2
Dilleniales	Dilleniaceae	*Hibbertia salicifolia*	*Hibbertia salicifolia*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Homalocalyx ericaeus*	*Homalocalyx ericaeus*	MAB	MAB-A	R	Actino	2
Malvales	Dipterocarpaceae	*Hopea pierrei*	*Hopea pierrei*	MAB	MAB-A	R	Actino	2
Malpighiales	Humiriaceae	*Humiria balsamifera*	*Humiria balsamifera*	MAB	MAB-A	R	Actino	2
Malpighiales	Hypericaceae	*Hypericum androsaemum*	*Hypericum androsatmum*	MAB	MAB-A	R	Actino	2
Malpighiales	Hypericaceae	*Hypericum tetrapterum*	*Hypericum quadrangulum*	MAB	MAB-A	R	Actino	2
Lamiales	Oleaceae	*Jasminum odoratissimum*	*Jasminum odoratissimum*	MAD	MAD-A	R	Actino	2
Lamiales	Oleaceae	*Jasminum pubigerum*	*Jasminum putbigerum*	MAD	MAD-A	R	Actino	2
Cornales	Loasaceae	*Kissenia spathulata*	*Kissenia spathulata*	MAB	MAB-A	R	Actino	2
Fabales	Fabaceae	*Laburnum vulgare*	*Laburnum vulgare*	MAD	MAD-0-CD	CD	Zygo	2
Myrtales	Combretaceae	*Laguncularia racemosa*	*Laguncularia racemosa*	MAB	MAB-A	R	Actino	2
Ericales	Ericaceae	*Kalmia buxifolium*	*Leiophyllum buxifolium*	MAD	MAD-A	R	Actino	2
Ericales	Ericaceae	*Leucopogon lanceolatus*	*Leucopogon lanceolatus*	MAB	MAB-A	R	Actino	2
Malpighiales	Linaceae	*Hugonia mystax*	*Linum austeiacum*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Nasa argemonoides*	*Loasa argemonoides*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Huidobria chilensis*	*Loasa chilensis*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Presliophytum incanum*	*Loasa incana*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Nasa urens*	*Loasa urens*	MAB	MAB-A	R	Actino	2
Gentianales	Loganiaceae	*Logania floribunda*	*Logania floribunda*	MAD	MAD-A	R	Actino	2
Myrtales	Combretaceae	*Lumnitzera coccinea*	*Lumnitzera coccinea*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Luxemburgia nobilis*	*Luxemburgia nobilis*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Mentzelia bartonioides*	*Mentzelia solierii*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Micromyrtus microphylla*	*Micromyrtus microphylla*	MAB	MAB-A	R	Actino	2
Caryophyllales	Montiaceae	*Montia fontana*	*Montia fonlaua*	MAD	MAD-A	R	Actino	2
Lamiales	Scrophulariaceae	*Myoporum sp*	*Myoporum sp*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Neckia serrata*	*Neckia serrata*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Ochna leucophloeos*	*Ochna leucophloeos*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Ouratea polygyna*	*Ouratea polygyna*	MAB	MAB-A	R	Actino	2
Solanales	Convolvulaceae	*Ipomoea hispida*	*Pharbitis hispida*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Pileanthus limacis*	*Pileanthus limacis*	MAB	MAB-A	R	Actino	2
Ericales	Ericaceae	*Pyrola sp*	*Pirola sp*	MAB	MAB-A	R	Actino	2

Asterales	Campanulaceae	*Platycodon grandiflorus*	*Platycodon grandiflorum*	MAB	MAB-A	R	Actino	2
Caryophyllales	Plumbaginaceae	*Plumbago europaea*	*Plumbago europaea*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Poecilandra retusa*	*Poecilandra retusa*	MAB	MAB-A	R	Actino	2
Malpighiales	Achariaceae	*Dasylepis blackii*	*Pyramidocarpus blackii*	MAD	MAD-A	R	Actino	2
Myrtales	Combretaceae	*Quisqualis indica*	*Quisqualis indica*	MAB	MAB-A	R	Actino	2
Ericales	Ericaceae	*Rhododendron luteum*	*Rhododendron flarum*	MAD	MAD-0-CD	CD	Zygo	2
Malpighiales	Violaceae	*Rinorea sp*	*Rinorea sp*	MAB	MAB-A	R	Actino	2
Saxifragales	Saxifragaceae	*Rodgersia podophylla*	*Rodgersia podophylla*	MAD	MAD-A	R	Actino	2
Sabiales	Sabiaceae	*Sabia lanceolata*	*Sabia lanceolata*	MAD	MAD-A	R	Actino	2
Malvales	Sarcolaenaceae	*Leptolaena sp*	*Sarcochluena grandiflora*	MAB	MAB-A	R	Actino	2
Malpighiales	Ochnaceae	*Sauvagesia tenella*	*Sauvagesia tenella*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Scyphanthus elegans*	*Scyphanthus elegans*	MAB	MAB-A	R	Actino	2
Sapindales	Meliaceae	*Entandrophragma angolense*	*Soymida febriftiga*	MAD	MAD-A	R	Actino	2
Caryophyllales	Plumbaginaceae	*Limonium vulgare*	*Statice limonium*	MAB	MAB-A	R	Actino	2
Malpighiales	Malpighiaceae	*Stigmaphyllon multilobum*	*Stigmatophyllon multilobum*	MAB	MAB-36-CD	CD	Zygo	2
Ericales	Styracaceae	*Styrax martii*	*Styrax martii*	MAB	MAB-A	R	Actino	2
Cornales	Loasaceae	*Eucnide rupestris*	*Sympetaleia rupestris*	MAB	MAB-A	R	Actino	2
Ericales	Pentaphylacaceae	*Ternstroemia sylvatica*	*Taonabo silvatica*	MAD	MAD-A	R	Actino	2
Myrtales	Myrtaceae	*Thryptomene mitchelliana*	*Thryptomene milchelliana*	MAB	MAB-A	R	Actino	2
Myrtales	Melastomataceae	*Tibouchina sp*	*Tibouchina sp*	MAB	MAB-A	R	Actino	2
Malpighiales	Passifloraceae	*Turnera ulmifolia*	*Turnera ulmifolia*	MAB	MAB-A	R	Actino	2
Sapindales	Sapindaceae	*Urvillea sp*	*Urvillea sp*	MAB	MAB-72-CV	CV	Zygo	2
Malpighiales	Humiriaceae	*Vantanea paniculata*	*Vantanea paniculata*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Verticordia densiflora*	*Verticordia densiflora*	MAB	MAB-A	R	Actino	2
Myrtales	Myrtaceae	*Verticordia picta*	*Verticordia picta*	MAB	MAB-A	R	Actino	2
Geraniales	Geraniaceae	*Viviania rosea*	*Viviania rosea*	MAD	MAD-A	R	Actino	2
Malpighiales	Ochnaceae	*Wallacea insignis*	*Wallacea insignis*	MAB	MAB-A	R	Actino	2
Canellales	Canellaceae	*Canella winterana*	*Wintrana canella*	MAB	MAB-A	R	Actino	2
Brassicales	Caricaceae	*Carica papaya*	*Carica papaya*	MAB	MAB-A	R	Actino	3, 2, 1
Rosales	Rosaceae	*Potentilla fruticosa*	*Potentilla fruticosa*	MAB	MAB-A	R	Actino	3, 2, 1
Brassicales	Tropaeolaceae	*Tropaeolum majus*	*Tropaeolum majus*	MAB	MAB-0-CV	CV	Zygo	3, 2, 1
Vitales	Vitaceae	*Ampelopsis hederacea*	*Ampelopsis hederacea*	MAD	MAD-A	R	Actino	3, 2
Lamiales	Acanthaceae	*Acanthus mollis*	*Acanthus mollis*	MAB	MAB-0-CV	CV	Zygo	3, 2
Ericales	Actinidiaceae	*Actinidia strigosa*	*Actinidia strigosa*	MAB	MAB-A	R	Actino	3, 2
Dipsacales	Adoxaceae	*Adoxa moschatellina*	*Adoxa moschatellina*	MAD	MAD-A	R	Actino	3, 2

Sapindales	Sapindaceae	*Aesculus hippocastanum*	*Aesculus hippocastanum*	MAB	MAB-72-CV	CV	Zygo	3, 2
Fabales	Fabaceae	*Amherstia nobilis*	*Amherstia nobilis*	MAD	MAD-0-CD	CD	Zygo	3, 2
Boraginales	Boraginaceae	*Anchusa officinalis*	*Anchusa officinalis*	MAB	MAB-A	R	Actino	3, 2
Gentianales	Apocynaceae	*Asclepias cornuti*	*Aselopias cornuti*	MAB	MAB-A	R	Actino	3, 2
Brassicales	Resedaceae	*Sesamoides clusii*	*Astrocarpus sesamoides*	MAB	MAB-0-CV	CV	Zygo	3, 2
Malpighiales	Linaceae	*Reinwardtia indica*	*Beinwardtia indiea*	MAB	MAB-A	R	Actino	3, 2
Malvales	Bixaceae	*Bixa orellana*	*Bixa orellana*	MAB	MAB-A	R	Actino	3, 2
Caryophyllales	Portulacaceae	*Calandrinia procumbens*	*Calandrinia procumbeus*	MAB	MAB-A	R	Actino	3, 2
Solanales	Convolvulaceae	*Calystegia sepium*	*Calystegia sepium*	MAB	MAB-A	R	Actino	3, 2
Asterales	Campanulaceae	*Campanula medium*	*Campanula medium*	MAB	MAB-A	R	Actino	3, 2
Fabales	Fabaceae	*Cercis siliquastrum*	*Cercis siliqnastrum*	MAD	MAD-0-CD	CD	Zygo	3, 2
Rosales	Rosaceae	*Chamaerhodos erecta*	*Chamaerhodos erecta*	MAB	MAB-A	R	Actino	3, 2
Fabales	Fabaceae	*Chorizema cordatum*	*Chorizema cordatum*	MAD	MAD-0-CD	CD	Zygo	3, 2
Ericales	Sapotaceae	*Chrysophyllum sp*	*Chrysophyllum sp*	MAB	MAB-A	R	Actino	3, 2
Malvales	Cistaceae	*Cistus acutifolius*	*Cistus acutifolins*	MAD	MAD-A	R	Actino	3, 2
Rosales	Rosaceae	*Potentilla palustre*	*Comarum palustre*	MAB	MAB-A	R	Actino	3, 2
Cucurbitales	Coriariaceae	*Coriaria myrtifolia*	*Coriaria myrtifolia*	MAB	MAB-A	R	Actino	3, 2
Malpighiales	Chrysobalanaceae	*Couepia macrophylla*	*Couepia macrophylla*	MAB	MAB-A	R	Actino	3, 2
Saxifragales	Crassulaceae	*Crassula lactea*	*Crassula lactea*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Crataegus sp*	*Crataegus sp*	MAB	MAB-A	R	Actino	3, 2
Cucurbitales	Cucurbitaceae	*Cucurbita pepo*	*Cucurbita pepo*	MAB	MAB-A	R	Actino	3, 2
Solanales	Convolvulaceae	*Cuscuta epithymum*	*Cuscuta epithymum*	MAB	MAB-A	R	Actino	3, 2
Solanales	Solanaceae	*Datura stramonium*	*Datura stramonium*	MAB	MAB-A	R	Actino	3, 2
Cornales	Hydrangeaceae	*Deutzia crenata*	*Deutzia crenata*	MAB	MAB-A	R	Actino	3, 2
Fabales	Fabaceae	*Dimorphandra sp*	*Diamorphaudra schott*	MAD	MAD-A	R	Actino	3, 2
Caryophyllales	Caryophyllaceae	*Dianthus plumarius*	*Dianthus plumarius*	MAB	MAB-A	R	Actino	3, 2
Caryophyllales	Droseraceae	*Dionaea muscipula*	*Dionaea muscipula*	MAB	MAB-A	R	Actino	3, 2
Ericales	Sapotaceae	*Sideroxylon salicifolia*	*Dipholis salicifolia*	MAB	MAB-A	R	Actino	3, 2
Boraginales	Boraginaceae	*Echium vulgare*	*Echium vulgare*	MAB	MAB-72-CV	CV	Zygo	3, 2
Lamiales	Acanthaceae	*Eranthemum nervosum*	*Eranthemum nervosum*	MAB	MAB-0-CV	CV	Zygo	3, 2
Geraniales	Geraniaceae	*Erodium cicutarium*	*Erodium cicutarium*	MAB	MAB-A	R	Actino	3, 2
Geraniales	Geraniaceae	*Geranium pratense*	*Geranium pratense*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Gillenia trifoliata*	*Gillenia trifoliata*	MAB	MAB-A	R	Actino	3, 2
Asterales	Goodeniaceae	*Goodenia ovata*	*Goodrnia ovata*	MAB	MAB-A	R	Actino	3, 2
Lamiales	Plantaginaceae	*Gratiola officinalis*	*Gratiola officinalis*	MAB	MAB-0-CV	CV	Zygo	3, 2

Cornales	Loasaceae	*Gronovia scandens*	*Gronovia scandens*	MAB	MAB-A	R	Actino	3, 2
Zygophyllales	Zygophyllaceae	*Guaiacum angustifolium*	*Guaiacum angutifolium*	MAB	MAB-A	R	Actino	3, 2
Malvales	Cistaceae	*Helianthemum sp*	*Helianthemum sp*	MAD	MAD-A	R	Actino	3, 2
Celastrales	Celastraceae	*Hippocratea schimperiana*	*Hippocratea schimperiana*	MAB	MAB-A	R	Actino	3, 2
Malpighiales	Chrysobalanaceae	*Hirtella hirsuta*	*Hirtella hirsuta*	MAB	MAB-A	R	Actino	3, 2
Malpighiales	Chrysobalanaceae	*Hirtella triandra*	*Hirtella triandra*	MAB	MAB-36-CD	CD	Zygo	3, 2
Solanales	Hydroleaceae	*Hydrolea spinosa*	*Hydrolea spinosa*	MAB	MAB-A	R	Actino	3, 2
Boraginales	Boraginaceae	*Hydrophyllum virginianum*	*Hydrophyllum virginianum*	MAB	MAB-A	R	Actino	3, 2
Ericales	Balsaminaceae	*Impatiens glandulifera*	*Impatiens glandilugora*	MAB	MAB-180-CD	CD	Zygo	3, 2
Ericales	Balsaminaceae	*Impatiens scabrida*	*Impatiens tricornis*	MAB	MAB-180-CD	CD	Zygo	3, 2
Myrtales	Onagraceae	*Ludwigia repens*	*Jussiaea repens*	MAB	MAB-A	R	Actino	3, 2
Malpighiales	Chrysobalanaceae	*Licania macrophylla*	*Licania macrophylla*	MAB	MAB-A	R	Actino	3, 2
Lamiales	Plantaginaceae	*Linaria vulgaris*	*Linaria vulgaris*	MAB	MAB-0-CV	CV	Zygo	3, 2
Dipsacales	Caprifoliaceae	*Linnaea borealis*	*Linnaea borealis*	MAB	MAB-0-CV	CV	Zygo	3, 2
Malpighiales	Linaceae	*Linum austriacum*	*Linum anstriacum*	MAB	MAB-A	R	Actino	3, 2
Asterales	Campanulaceae	*Lobelia fulgens*	*Lobelia fulgens*	MAD	MAD-180-CV	CV	Zygo	3, 2
Myrtales	Onagraceae	*Lopezia racemosa*	*Lopezia eacemosa*	MAB	MAB-0-CV	CV	Zygo	3, 2
Myrtales	Onagraceae	*Lopezia sp*	*Lopezia eacemosa*	MAB	MAB-0-CV	CV	Zygo	3, 2
Rosales	Rosaceae	*Mespilus germanica*	*Mespilus gemaniea*	MAB	MAB-A	R	Actino	3, 2
Sapindales	Rutaceae	*Ertela trifolia*	*Monnieria trifolia*	MAB	MAB-36-CD	CD	Zygo	3, 2
Geraniales	Geraniaceae	*Monsonia biflora*	*Monsinia bifora*	MAB	MAB-A	R	Actino	3, 2
Myrtales	Myrtaceae	*Myrtus communis*	*Myrtus communis*	MAB	MAB-A	R	Actino	3, 2
Ericales	Theaceae	*Gordonia lasianthus*	*Nach herbarmateria*	MAB	MAB-A	R	Actino	3, 2
Solanales	Solanaceae	*Nicandra physalodes*	*Nicandra physaloides*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Oemleria cerasiformis*	*Nuttallia cerasiformis*	MAB	MAB-A	R	Actino	3, 2
Oxalidales	Oxalidaceae	*Oxalis acetosella*	*Oxalis acetosella*	MAB	MAB-A	R	Actino	3, 2
Oxalidales	Oxalidaceae	*Oxalis corniculata*	*Oxalis corniculata*	MAB	MAB-A	R	Actino	3, 2
Malpighiales	Chrysobalanaceae	*Parinari sp*	*Parinarium gardneri*	MAB	MAB-A	R	Actino	3, 2
Fabales	Fabaceae	*Parkia africana*	*Parkia africana*	MAB	MAB-A	R	Actino	3, 2
Sapindales	Nitrariaceae	*Peganum harmala*	*Peganum harmala*	MAB	MAB-A	R	Actino	3, 2
Geraniales	Geraniaceae	*Pelargonium zonale*	*Pelargouium zonale*	MAB	MAB-0-CV	CV	Zygo	3, 2
Solanales	Solanaceae	*Petunia nyctaginiflora*	*Petunia nyctaginiflora*	MAB	MAB-36-CD	CD	Zygo	3, 2
Asterales	Campanulaceae	*Phyteuma spicatum*	*Phytouma spicatum*	MAB	MAB-A	R	Actino	3, 2
Lamiales	Lentibulariaceae	*Pinguicula alpina*	*Pinguicula alpina*	MAB	MAB-0-CV	CV	Zygo	3, 2
Rosales	Rosaceae	*Pyrus communis*	*Pirns communis*	MAB	MAB-A	R	Actino	3, 2

Apiales	Pittosporaceae	*Pittosporum undulatum*	*Pirrosporum undularum*	MAB	MAB-A	R	Actino	3, 2
Ericales	Polemoniaceae	*Polemonium caeruleum*	*Polemonium coeruleum*	MAB	MAB-A	R	Actino	3, 2
Caryophyllales	Portulacaceae	*Portulaca oleracea*	*Portulaca oleracea*	MAB	MAB-A	R	Actino	3, 2
Ericales	Primulaceae	*Primula acaulis*	*Primula acaulis*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Prunus padus*	*Prunus padus*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Prunus virginiana*	*Prunus virginiana*	MAB	MAB-A	R	Actino	3, 2
Dipsacales	Caprifoliaceae	*Pterocephalus palaestinus*	*Pterocephalus palaesonus*	MAB	MAB-0-CV	CV	Zygo	3, 2
Fabales	Quillajaceae	*Quillaja brasiliensis*	*Quillaja brasiliensis*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Rhaphiolepis sp*	*Raphiolepis sp*	MAB	MAB-A	R	Actino	3, 2
Brassicales	Resedaceae	*Randonia sp*	*Reaeda alba*	MAB	MAB-A	R	Actino	3, 2
Brassicales	Resedaceae	*Reseda alba*	*Reaeda alba*	MAB	MAB-A	R	Actino	3, 2
Saxifragales	Grossulariaceae	*Ribes alpinum*	*Ribes alpinum*	MAB	MAB-A	R	Actino	3, 2
Saxifragales	Grossulariaceae	*Ribes sanguineum*	*Ribes sanguineum*	MAB	MAB-A	R	Actino	3, 2
Sapindales	Rutaceae	*Ruta graveolens*	*Ruta graveolens*	MAB	MAB-A	R	Actino	3, 2
Solanales	Solanaceae	*Salpiglossis sinuata*	*Salpiglossis sinnata*	MAB	MAB-36-CD	CD	Zygo	3, 2
Myrtales	Vochysiaceae	*Salvertia convallariaeodora*	*Salvertia convallariodore*	MAB	MAB-72-CV	CV	Zygo	3, 2
Dipsacales	Adoxaceae	*Sambucus canadensis*	*Sambucus canadensis*	MAB	MAB-A	R	Actino	3, 2
Dipsacales	Adoxaceae	*Sambucus ebulus*	*Sambucus ebulum*	MAB	MAB-A	R	Actino	3, 2
Saxifragales	Saxifragaceae	*Saxifraga granulata*	*Saxifraga granulate*	MAB	MAB-A	R	Actino	3, 2
Saxifragales	Saxifragaceae	*Saxifraga stolonifera*	*Saxifraga sarmentosa*	MAB	MAB-36-CD	CD	Zygo	3, 2
Dipsacales	Caprifoliaceae	*Scabiosa atropurpurea*	*Scabiosa atropurpurea*	MAB	MAB-0-CV	CV	Zygo	3, 2
Solanales	Solanaceae	*Schizanthus retusus*	*Schizanthus retusus*	MAB	MAB-36-CD	CD	Zygo	3, 2
Rosales	Rosaceae	*Sibbaldia cuneata*	*Sibbaldia cuneata*	MAB	MAB-A	R	Actino	3, 2
Ericales	Sapotaceae	*Sideroxylon sp*	*Sideroxylon sp*	MAB	MAB-A	R	Actino	3, 2
Caryophyllales	Caryophyllaceae	*Silene inflata*	*Silene inflata*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Sorbus domestica*	*Sorbus domestica*	MAB	MAB-A	R	Actino	3, 2
Asterales	Campanulaceae	*Legousia speculum*	*Specularia speeulum*	MAB	MAB-A	R	Actino	3, 2
Caryophyllales	Caryophyllaceae	*Spergula arvensis*	*Spergula arvensis*	MAB	MAB-A	R	Actino	3, 2
Rosales	Rosaceae	*Spiraea hypericifolia*	*Spiraea hypericifolia*	MAB	MAB-A	R	Actino	3, 2
Celastrales	Celastraceae	*Stackhousia monogyna*	*Stackhousia monogyna*	MAB	MAB-A	R	Actino	3, 2
Crossosomatales	Staphyleaceae	*Staphylea trifoliata*	*Staphylea trifoliata*	MAB	MAB-A	R	Actino	3, 2
Caryophyllales	Plumbaginaceae	*Limonium latifolia*	*Statice latifolia*	MAB	MAB-A	R	Actino	3, 2
Asterales	Stylidiaceae	*Stylidium adnatum*	*Stylidium adnatum*	MAB	MAB-0-CV	CV	Zygo	3, 2
Dipsacales	Caprifoliaceae	*Symphoricarpos racemosus*	*Symphorica racomosa*	MAB	MAB-A	R	Actino	3, 2
Dilleniales	Dilleniaceae	*Tetracera volubilis*	*Tetracera volubilis*	MAB	MAB-A	R	Actino	3, 2

Zygophyllales	Zygophyllaceae	*Tribulus terrestris*	*Tribulus terrestris*	MAD	MAD-A	R	Actino	3, 2
Malpighiales	Trigoniaceae	*Trigonia simplex*	*Trigionia simplex*	MAB	MAB-36-CD	CD	Zygo	3, 2
Lamiales	Lentibulariaceae	*Utricularia vulgaris*	*Utricularia vulgaris*	MAB	MAB-0-CV	CV	Zygo	3, 2
Lamiales	Scrophulariaceae	*Verbascum nigrum*	*Verbascum nigrum*	MAB	MAB-A	R	Actino	3, 2
Lamiales	Plantaginaceae	*Veronica chamaedrys*	*Veroniaca chamaedrys*	MAB	MAB-0-CV	CV	Zygo	3, 2
Dipsacales	Adoxaceae	*Viburnum lantana*	*Viburuum lantana*	MAB	MAB-A	R	Actino	3, 2
Fabales	Fabaceae	*Vicia faba*	*Vicia faba*	MAD	MAD-0-CD	CD	Zygo	3, 2
Malpighiales	Violaceae	*Viola sp*	*Viola sp*	MAB	MAB-0-CV	CV	Zygo	3, 2
Caryophyllales	Caryophyllaceae	*Silene vulgaris*	*Viscaria vulgaris*	MAB	MAB-A	R	Actino	3, 2
Ericales	Pentaphylacaceae	*Visnea mocanera*	*Visnea mocanera*	MAB	MAB-A	R	Actino	3, 2
Zygophyllales	Zygophyllaceae	*Zygophyllum fabago*	*Zygophyllum fabago*	MAB	MAB-A	R	Actino	3, 2
Sapindales	Sapindaceae	*Acer pseudoplatanus*	*Acer pseudoplatanus*	MAB	MAB-A	R	Actino	3
Fabales	Fabaceae	*Aldina latifolia*	*Aldina latifolia*	MAD	MAD-A	R	Actino	3
Caryophyllales	Droseraceae	*Aldrovanda vesiculosa*	*Aldrovandia vesieulosa*	MAD	MAD-A	R	Actino	3
Malvales	Malvaceae	*Althaea rosea*	*Althaea rosen*	MAD	MAD-A	R	Actino	3
Sapindales	Anacardiaceae	*Anacardium occidentale*	*Anacurdium occidentale*	MAB	MAB-36-CD	CD	Zygo	3
Ranunculales	Ranunculaceae	*Aquilegia vulgaris*	*Aquilegia vulagaris*	MAB	MAB-A	R	Actino	3
Apiales	Araliaceae	*Aralia spinosa*	*Aralia spinisn*	MAB	MAB-A	R	Actino	3
Ericales	Ericaceae	*Rhododendron ponticum*	*Azalea pontiea*	MAD	MAD-A	R	Actino	3
Caryophyllales	Basellaceae	*Basella rubra*	*Basella rubra*	MAB	MAB-A	R	Actino	3
Myrtales	Myrtaceae	*Beaufortia decussata*	*Beaufortia decussata*	MAB	MAB-A	R	Actino	3
Malpighiales	Elatinaceae	*Bergia ammannioides*	*Bergia ammannioides*	MAB	MAB-A	R	Actino	3
Sapindales	Anacardiaceae	*Cotinus coggygria*	*Bhus cotinus*	MAB	MAB-A	R	Actino	3
Lamiales	Bignoniaceae	*Bignonia unguis*	*Bignonia unguis*	MAB	MAB-0-CV	CV	Zygo	3
Cornales	Loasaceae	*Caiophora lateritia*	*Cajophora lateritia*	MAB	MAB-A	R	Actino	3
Caryophyllales	Portulacaceae	*Cistanthe grandiflora*	*Calandrinia speciosa*	MAB	MAB-A	R	Actino	3
Myrtales	Myrtaceae	*Callistemon salignus*	*Callistemon salignus*	MAB	MAB-A	R	Actino	3
Malpighiales	Malpighiaceae	*Camarea triphylla*	*Camarea triphylla*	MAB	MAB-36-CD	CD	Zygo	3
Asterales	Asteraceae	*Carduus crispus*	*Carduus crispus*	MAB	MAB-A	R	Actino	3
Fabales	Fabaceae	*Cassia caroliniana*	*Cassia caroliniana*	MAD	MAD-0-CD	CD	Zygo	3
Sapindales	Rutaceae	*Citrus aurantium*	*Citrus aurantium*	MAB	MAB-A	R	Actino	3
Malpighiales	Clusiaceae	*Clusia sp*	*Clusia sp*	MAD	MAD-A	R	Actino	3
Sapindales	Rutaceae	*Coleonema album*	*Coleonema album*	MAB	MAB-A	R	Actino	3
Myrtales	Combretaceae	*Combretum sp*	*Combretum sp*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Corchorus siliquosus*	*Corehorus siliquosns*	MAB	MAB-A	R	Actino	3

Fabales	Fabaceae	*Cytisus laburnum*	*Cytisus laburnum*	MAD	MAD-0-CD	CD	Zygo	3
Ericales	Sarraceniaceae	*Darlingtonia californica*	*Darlingtnia californica*	MAB	MAB-A	R	Actino	3
Myrtales	Lythraceae	*Decodon aquaticus*	*Docodon aquaticus*	MAB	MAB-A	R	Actino	3
Caryophyllales	Droseraceae	*Drosera rotundifolia*	*Drosera ritnndifolia*	MAB	MAB-A	R	Actino	3
Saxifragales	Crassulaceae	*Echeveria gibbiflora*	*Echeveria gibbiflora*	MAB	MAB-A	R	Actino	3
Apiales	Apiaceae	*Eryngium maritimum*	*Eryngium maritimum*	MAB	MAB-A	R	Actino	3
Escalloniales	Escalloniaceae	*Escallonia floribunda*	*Escallonia floribunda*	MAB	MAB-A	R	Actino	3
Sapindales	Rutaceae	*Galipea jasminiflora*	*Galipea jasminiflora*	MAB	MAB-A	R	Actino	3
Gentianales	Apocynaceae	*Geissospermum vellosii*	*Geissospermum vellosii*	MAB	MAB-A	R	Actino	3
Gentianales	Gentianaceae	*Gentiana verna*	*Gentiana verna*	MAB	MAB-A	R	Actino	3
Lamiales	Gesneriaceae	*Sinningia sp*	*Gesnera pendulina*	MAB	MAB-0-CV	CV	Zygo	3
Lamiales	Plantaginaceae	*Globularia nudicaulis*	*Globularia nudieaulis*	MAB	MAB-0-CV	CV	Zygo	3
Malvales	Malvaceae	*Glossostemon sp*	*Glossostemon spinesceus*	MAB	MAB-A	R	Actino	3
Sapindales	Rutaceae	*Glycosmis pentaphylla*	*Glycismis pentaphylla*	MAB	MAB-A	R	Actino	3
Caryophyllales	Frankeniaceae	*Frankenia pulverulenta*	*Grundriss desselben*	MAB	MAB-A	R	Actino	3
Caryophyllales	Nyctaginaceae	*Mirabilis jalapa*	*Grundriss sp*	MAB	MAB-A	R	Actino	3
Apiales	Araliaceae	*Hedera helix*	*Hedera helix*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Hermannia denudata*	*Hermannia denndata*	MAB	MAB-A	R	Actino	3
Saxifragales	Saxifragaceae	*Heuchera americana*	*Heuchera americana*	MAB	MAB-A	R	Actino	3
Dilleniales	Dilleniaceae	*Hibbertia microphylla*	*Hibbertia microphylla*	MAB	MAB-A	R	Actino	3
Malpighiales	Malpighiaceae	*Hiptage madablota*	*Hiptage modablota*	MAB	MAB-36-CD	CD	Zygo	3
Solanales	Solanaceae	*Hyoscyamus albus*	*Hyoseyamus albus*	MAB	MAB-36-CD	CD	Zygo	3
Malpighiales	Hypericaceae	*Triadenum virginicum*	*Hypericum virginicum*	MAB	MAB-A	R	Actino	3
Solanales	Convolvulaceae	*Ipomoea purpurea*	*Ipomoea purpurea*	MAB	MAB-A	R	Actino	3
Ericales	Primulaceae	*Jacquinia armillaris*	*Jacquinia armillaris*	MAB	MAB-A	R	Actino	3
Malpighiales	Malpighiaceae	*Janusia amazonica*	*Jannsia amazoniea*	MAB	MAB-A	R	Actino	3
Lamiales	Lamiaceae	*Lamium album*	*Lamium album*	MAB	MAB-0-CV	CV	Zygo	3
Lamiales	Orobanchaceae	*Lathraea squamaria*	*Lathraea squamaria*	MAB	MAB-0-CV	CV	Zygo	3
Ericales	Ericaceae	*Leucopogon cunninghamii*	*Leucopogon cunnfughamii*	MAB	MAB-A	R	Actino	3
Brassicales	Limnanthaceae	*Limnanthes douglasii*	*Limnanthes douglasii*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Luehea sp*	*Luhea sp*	MAB	MAB-A	R	Actino	3
Malpighiales	Malpighiaceae	*Malpighia macrophylla*	*Malpighia macrophylla*	MAB	MAB-A	R	Actino	3
Celastrales	Celastraceae	*Maytenus chilensis*	*Maytenus chilensis*	MAB	MAB-A	R	Actino	3
Sapindales	Meliaceae	*Melia azedarach*	*Melia azedarach*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Melochia pyramidata*	*Melochia pyramidata*	MAB	MAB-A	R	Actino	3

Cornales	Loasaceae	*Mentzelia lindleyi*	*Mentzelia lindleyi*	MAB	MAB-A	R	Actino	3
Saxifragales	Saxifragaceae	*Mitella pentandra*	*Mitellopsis pentsndra*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Mollia sp*	*Molllia sp*	MAB	MAB-A	R	Actino	3
Ericales	Ericaceae	*Monotropa hypopitys*	*Monotropa hypopitys*	MAB	MAB-A	R	Actino	3
Lamiales	Scrophulariaceae	*Myoporum parvifolium*	*Myoporum parvifolium*	MAB	MAB-A	R	Actino	3
Caryophyllales	Tamaricaceae	*Myricaria germanica*	*Myricaria germanica*	MAB	MAB-A	R	Actino	3
Ranunculales	Ranunculaceae	*Nigella orientale*	*Nigellastrum orientale*	MAB	MAB-A	R	Actino	3
Ericales	Marcgraviaceae	*Norantea sp*	*Norantea sp*	MAB	MAB-A	R	Actino	3
Lamiales	Orobanchaceae	*Orobanche flava*	*Orobanche flava*	MAB	MAB-0-CV	CV	Zygo	3
Saxifragales	Paeoniaceae	*Paeonia officinalis*	*Paeonia officinalis*	MAB	MAB-A	R	Actino	3
Malpighiales	Passifloraceae	*Passiflora sp*	*Passiflora sp*	MAB	MAB-A	R	Actino	3
Ericales	Ericaceae	*Pyrola rotundifolia*	*Pirola rotundifolia*	MAB	MAB-A	R	Actino	3
Malpighiales	Clusiaceae	*Platonia insignis*	*Platonia insignismart*	MAB	MAB-A	R	Actino	3
Oxalidales	Elaeocarpaceae	*Platytheca verticillata*	*Platytheca verticillate*	MAB	MAB-A	R	Actino	3
Caryophyllales	Plumbaginaceae	*Plumbago larpentae*	*Plumbago larpentae*	MAD	MAD-A	R	Actino	3
Caryophyllales	Portulacaceae	*Portulaca grandiflora*	*Portulaca grandiflora*	MAB	MAB-A	R	Actino	3
Ranunculales	Ranunculaceae	*Ranunculus acris*	*Ranunculus acris*	MAB	MAB-A	R	Actino	3
Malpighiales	Clusiaceae	*Clusia comans*	*Renggeria comans*	MAB	MAB-A	R	Actino	3
Rosales	Rhamnaceae	*Rhamnus frangula*	*Rhamnus frangula*	MAB	MAB-A	R	Actino	3
Ericales	Ericaceae	*Rhododendron hirsutum*	*Rhododendron hirsutum*	MAD	MAD-A	R	Actino	3
Rosales	Rosaceae	*Rosa tomentosa*	*Rosa tomentosa*	MAB	MAB-A	R	Actino	3
Gentianales	Rubiaceae	*Rubia tinctorum*	*Rubia tinctorum*	MAB	MAB-A	R	Actino	3
Ericales	Sarraceniaceae	*Sarracenia purpurea*	*Sarracenia purpurea*	MAB	MAB-A	R	Actino	3
Malpighiales	Ochnaceae	*Sauvagesia sprengelii*	*Sauvagesia sprengelii*	MAB	MAB-A	R	Actino	3
Sapindales	Anacardiaceae	*Schinus molle*	*Schinus molle*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Sidalcea diploscypha*	*Sidalcea diploscypha*	MAB	MAB-A	R	Actino	3
Ericales	Marcgraviaceae	*Ruyschia amazonica*	*Sourubea amazonica*	MAB	MAB-A	R	Actino	3
Caryophyllales	Caryophyllaceae	*Stellaria media*	*Stellaria media*	MAB	MAB-A	R	Actino	3
Malpighiales	Malpighiaceae	*Stigmaphyllon sp*	*Stigmaphyllon sp*	MAB	MAB-36-CD	CD	Zygo	3
Gentianales	Loganiaceae	*Strychnos nuxvomica*	*Strychnos nux-vomica*	MAB	MAB-A	R	Actino	3
Lamiales	Lamiaceae	*Salvia officinalis*	*Sulvia officinalis*	MAB	MAB-0-CV	CV	Zygo	3
Ericales	Pentaphylacaceae	*Ternstroemia sp*	*Ternstroemia silvestris*	MAD	MAD-A	R	Actino	3
Malvales	Malvaceae	*Tilia americana*	*Tilia amerieana*	MAB	MAB-A	R	Actino	3
Malvales	Malvaceae	*Tilia platyphyllos*	*Tilia grandifolia*	MAB	MAB-A	R	Actino	3
Malpighiales	Passifloraceae	*Turnera sp*	*Turnera sp*	MAB	MAB-A	R	Actino	3

Ericales	Ericaceae	*Vaccinium vitis-idaea*	*Vaccinium vitis*	MAB	MAB-A	R	Actino	3
Gentianales	Apocynaceae	*Vinca minor*	*Vinca minor*	MAB	MAB-A	R	Actino	3
Malpighiales	Hypericaceae	*Vismia cayennensis*	*Vismia cayennensis*	MAB	MAB-A	R	Actino	3
Rosales	Rhamnaceae	*Ziziphus vulgaris*	*Zizyphus vulgaris*	MAB	MAB-A	R	Actino	3

Appendix table 2. The references to solve the relationships in the phylogenetic tree.

Clade	Referance
Saxifragales	(Moorea, Soltisb, Bellc, Burleighd, & Soltis, 2010)
Malpighiales	(Xi et al., 2012)
Malvales	(Alverson et al., 1998)
Ericales	(Schonenberger, Anderberg, & Sytsma, 2005)
Pentaphylacaceae	(Luna & Ochoterena, 2004)
Balsaminaceae	(Janssens et al., 2006)
Fabaceae	1- (Bruneau et al., 2013) 2- (Sinou, Forest, Lewis, & Bruneaua, 2009)
Rutaceae	1- (Groppo, Pirani, Salatino, Blanco, & Kallunki, 2008) 2- (Morton & Kallunki, 1993)
Ochnaceae	(Schneider et al., 2014)
Aricaceae	1- (Kron, 1996) 2- (Goetsch, Eckert, & Hall, 2005)
Myrtaceae	(Wilson, Heslewood, Lam, & Quinn, 2004)
Lamiaceae	(Bendiksby, Thorbek, Scheen, Lindqvist, & Ryding, 2011)
Acanthaceae	1- (McDade, Masta, Moody, & Waters, 2000) 2- (McDade & Moody, 1999)
Hyperiacaceae	(Park & Kim, 2004)
Dilleniaceae	(Horn, 2009)
Loasaceae	1- (Hufford, Mcmahon, Sherwood, Reeves, & Chase, 2003) 2- (Moody, Hufford, Soltis, & Soltis, 2001)
Portulacaceae	(Applequist & Wallace, 2001)
Caryophyllaceae	(Fior, Karis, Casazza, Minuto, & Sala, 2006)
Grossulariaceae	(Schultheis & Donoghue, 2004)
Anacardiaceae	(Wannan, 2006)
Melastomataceae	(Clausing & Renner, 2001)
Combretaceae	(Maurin, Chase, Jordaan, & Van Der Bank, 2010)
Melianthaceae	(Linder, Dlamini, Henning, & Verboom, 2006)
Primulaceae	(Kallersjo, Bergqvist, & Anderberg, 2000)
Araliaceae	(Wen, Plunkett, Mitchell, & Wagstaff, 2001)

Convolvulaceae	(Neyland, 2001)
Ranunculaceae	(Hoot, 1991)
Saxifragaceae	(Deng et al., 2015)
Adoxaceae	(W. H. Zhang, Chen, Li, Chen, & Tang, 2003)
Caprifoliaceae	(W. H. Zhang et al., 2003)
Boraginaceae	(Nazaire & Hufford, 2012)
Solanaceae	(Olmstead et al., 2008)
Zygophyllaceae	(Sheahan & Chase, 2000)
Chrysobalanaceae	(Yakandawala, Morton, & Prance, 2010)
Malpighiaceae	(Davis & Anderson, 2010)
Linaceae	(Mcdill & Simpson, 2010)
Clusiaceae	(Ruhfel, Stevens, & Davis, 2013)
Geraniaceae	(Price & Palmer, 1993)
Campanulaceae	(Eddie, Shulkina, Goskin, Haberle, & Jansen, 2003)
Apocynaceae	(Potgieter & Albert, 2001)
Loganiaceae	(Backlund, Oxelman, & Bremer, 2000)
Hydrangeaceae	(Hufford, Moody, & Soltis, 2001)
Plumbaginaceae	(Lledo, Crespo, Fay, & Chase, 2005)
Droseraceae	(Rivadavia, Kondo, Kato, & Hasebe, 2003)
Plantaginaceae	(Albach, Meudt, & Oxelman, 2005)
Scrophulariaceae	(Oxelman, Kornhall, Olmstead, & Bremer, 2005)
Celasreaceae	(Simmons, Savolainen, Clevinger, Archer, & Davis, 2001)
Rosaceae	1- (Potter et al., 2007) 2- (Campbell, Evans, Morgan, Dickinson, & Arsenault, 2007)
Resedaceae	(Martin-Bravo et al., 2007)

Appendix table 3. Molecular markers from gene bank used to resolve the phylogenetic tree.

Family Genus	Genus	Tree species	Genbank species	MARKERS													
				matK	ITS	rbcL	trnL	ndhF	trnL-F	rps4	psbA-trnH	trnH	rpl16	18S	18S+28S+ITS	18S+ITS	trnK
Malvaceae	abutilon	Abutilon megapotamicum	Abutilon theophrasti	HM850990.1	DQ287984.1	HM849734.1			HQ696727.1								
Malvaceae	althaea	Altheae rosea	Althaea officinalis	JN894570.1	AF303026.1	KM360627.1			EF679770.1								
Malvaceae	ayenia	Ayenia cordobensis	Ayenia micrantha	JQ589283.1		JQ594193.1											
Malvaceae	ceiba	Criba pentandra	Ceiba pentandra	GQ981960.1	HQ658386.1	JX987572.1			HQ696753.1								
Malvaceae	corchorus	Corchorus siliquosus	Corchorus siliquosus	JQ693592.1	FJ527604.1				JQ625349.1								
Malvaceae	glossostemon	Glossostemon sp	Glossostemon bruguieri					AF287922.1									
Malvaceae	hermannia	Hermannia denudata	Hermannia verticillata					AF287919.1									
Malvaceae	luhea	Luhea sp	Luehea speciosa	JQ589348.1		JQ594266.1											
Malvaceae	melochia	Melochia pyramidata	Melochia tomentosa	JQ589306.1		KJ082418.1											
Malvaceae	mollia	Mollia sp	Mollia speciosa					JF809667.1									
Malvaceae	sidalcea	Sidalcea diploscypha	Sidalcea diploscypha		AJ849680.1												
Malvaceae	theobroma	Theobroma cacao	Theobroma cacao	HM488452.1	AY074729.1	AF022125.1		AF287916.1									
Malvaceae	tillia	Tilia americana	Tilia americana	HQ593469.1	KF445422.1	AF022127.1											
Malvaceae	tillia	Tilia europaea	Tilia europaea	FJ395		KM36											

				445.1		1014.1											
Malvaceae	tillia	Tilia grandifolia	sy: Tilia platyphyllos	JN894341.1	KF445429.1	KP088889.1											
Outgroup-Neuradaceae	Neurada	Neurada procumbens	Neurada procumbens		KJ004316.1	U06814.1		EU002245.1									
Outgroup-Resedaceae	Randonia	Randonia sp	Randonia africana		DQ987084.1	GQ891211.1			DQ986982.1								
Myrtaceae	callistemon	Callistemon salignus	Callistemon salignus	KM065378.1											KM064943.1		
Myrtaceae	callistemon	Callistemon lanceolatus	Callistemon lanceolatus		JX856559.1	JX856671.1											
Myrtaceae	callistemon	Callistemon citrinus	Callistemon citrinus	KM065347.1	JX856557.1	JX856668.1								KM064753.1			
Myrtaceae	beaufortia	Beaufortia decussata	Beaufortia orbifolia	AY521530.1				AY498771.1								AF048888.1	
Myrtaceae	verticordia	Verticodia picta	Verticordia staminosa	AY259828.1													
Myrtaceae	verticordia	Verticodia densiflora	Verticordia staminosa	AY259828.1													
Myrtaceae	homalocalyx	Homalocalyx ericaeus	Homalocalyx aurea	AF489398.1	HM160106.1			AY498785.1									
Myrtaceae	chamelaucium	Chamelaucium ciliatum	Chamelaucium uncinatum	KM065367.1	KM065058.1									EU850639.1			
Myrtaceae	micromyrtus	Micromyrtus microphylla	Micromyrtus ciliata	KM065333.1	KM064940.1												
Myrtaceae	myrtus	Myrtus communis	Myrtus communis	KM065333.1	JQ740194.1	HM850194.1		AF215593.1						JF304901.1			
Myrtaceae	calytrix	Calytrix tetragona	Calytrix tetragona	KM065336.1	KM064975.1	AF489340.1		AY498776.1									
Myrtaceae	darwinia	Darwinia oederoides	Darwinia vestita	KM065194.	KM065008.												

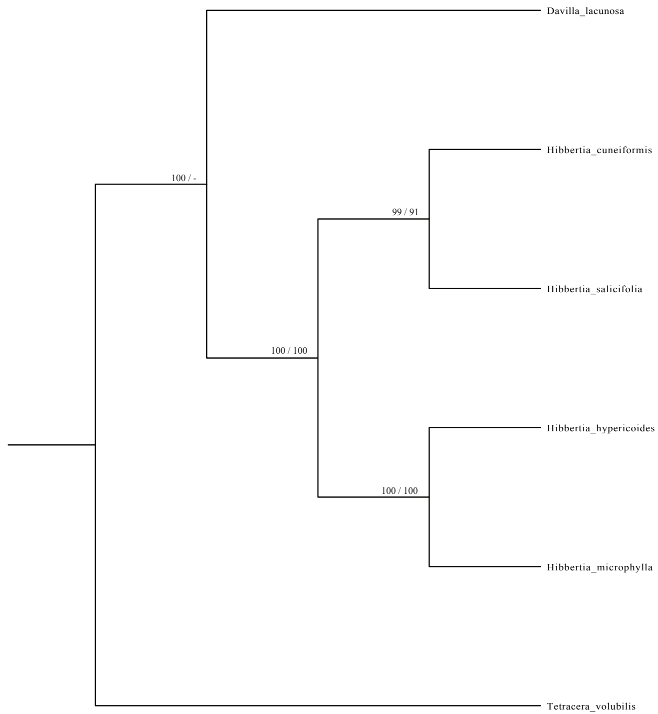
				1	1												
Myrtaceae	pileanthus	Pileanthus limacis	Pileanthus filifolius	EF581210.1		EF581246.1		EF581231.1						EF581195.1			
Myrtaceae	thryptomene	Thryptomene mitchelliana	Thryptomene calycina	KM065120.1	KM064883.1												
Outgroup-Melastomataceae	Medinilla	Medinilla magnifica	Medinilla septentrionalis	KP093855.1		KP094795.1										KP093025.1	
Outgroup-Onagraceae	Ludwigia	Ludwigia repens	Ludwigia repens	KJ772917.1		KJ773653.1											
Acanthaceae	eranthemum	Eranthemum nervosum	Eranthemum nervosum			JQ933327.1											
Acanthaceae	acanthus	Acanthus mollis	Acanthus mollis	HM850911.1	DQ028416.1	HM849737.1			KM888789.1								
Acanthaceae	odontonema	Odontonema strictum	Odontonema tubiforme	JQ586417.1	AF169748.1	JQ590073.1											
Outgroup-Bignoniaceae	bignonia	Bignonia unguis	Bignonia aequin octialis	KJ593845.1		KJ594211.1			FJ870018.1								
Outgroup-Plantaginaceae	Veronica	Veronica chamaedrys	Veronica chamaedrys	KJ746160.1	AF313003.1	KJ746275.1			KJ646890.1								
Lamiaceae	lamium	Lamium album	Lamium album	AJ429332.1		KM360840.1											
Lamiaceae	callicarpa	Callicarpa farinosa	Callicarpa farinosa			JF739109.1											
Lamiaceae	clerodendrum	Clerodendrum perasites	Clerodendrum trichotomum	HQ384492.1		HQ384865.1											
Lamiaceae	salvia	Salvia officinalis	Salvia officinalis	KC473367.1		AY570431.1											
Outgroup-Orobanchaceae	lathraea	Lathraea squamaria	Lathraea squamaria	KC542164.1		JN890561.1											
Outgroup-Scrophulariaceae	myoporum	Myoporum parvifolium	Myoporum mauritianum	HQ384532.1		L36445.1											

Outgroup-Acanthaceae	odontonema	Odontonema strictum	Odontonema tubiforme	JQ586417.1													
Sapindaceae	urvillea	Urvillea sp	Urvillea ulmacea	EU720655.1	EU720499.1												
Sapindaceae	sarjania	Serjania glabrata	Serjania glabrata	EU720703.1	EU720557.1	GU935454.1											
Sapindaceae	aesculus	Aesculus hippocastanum	Aesculus pavia	AJ581450.1	EU687669.1												
Sapindaceae	acer	Acer pseudoplatanus	Acer pseudoplatanus	KJ204427.1		HM849739.1											
Outgroup-Anacardiaceae	rhus	Rhus cotinus	Cotinus coggygria	HE966907.1		AY510148.1											
Outgroup-Nitraariaceae	peganum	Peganum harmala	Peganum harm ala	AY177667.1	KP087776.1												
Tilia	tillia	Tilia americana	Tilia americana	HQ593469.1	KF445422.1	AF022127.1		AF111760.1									
Tilia	tillia	Tilia europaea	Tilia europaea	FJ395445.1		KM361014.1											
Tilia	tillia	Tilia grandifolia	sy: Tilia platyphyllos	JN894341.1	KF445429.1	KP088889.1											
Outgroup-Neuradaceae	Neurada	Neurada procumbens	Neurada procumbens		KJ004316.1	U06814.1		EU002245.1									
Outgroup-Malvaceae	theobroma	Theobroma cacao	Theobroma cacao	HM488452.1	AY074729.1	AF022125.1		AF287916.1									
Bauhinia	Bauhinia	Bauhinia anguina	sy: Bauhinia scandens	JN881423.1	AY258408.1												
Bauhinia	Bauhinia	Bauhinia forficata	Bauhinia forficata	JN881365.1					FJ801113.1								JN881365.1
Bauhinia	Bauhinia	Bauhinia krugii	sy: Bauhinia monandra	JN881378.1					FJ801127.1								JN881378.1

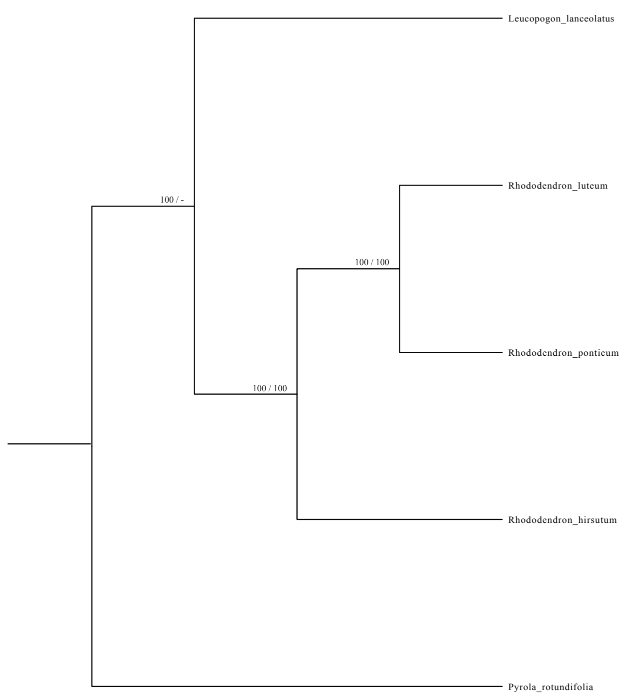
Bauhinia	Bauhinia	Bauhinia pauletia	Bauhinia pauletia	JN881381.1					FJ801087.1								
Bauhinia	Bauhinia	Bauhinia purpurea	Bauhinia purpurea	JN881391.1	JX856407.1	AF387980.1			FJ801075.1								
Bauhinia	Bauhinia	Bauhinia tomentosa	Bauhinia tomentosa	AY386893.1	AY126643.1	JX572328.1			FJ801088.1								JN881403.1
Outgroup-Fabaceae	Outgroup for Bauhinia	Amherstia nobilis	Amherstia nobilis	AF542601.1		AM234234.1			AF549295.2								EU361849.1
Outgroup-Fabaceae	Outgroup for Bauhinia	Afzelia quanzensis	Afzelia quanzensis	JF270629.1		JX572247.1											EU361848.1
Zanthoxylum	Fagara	Fagara caribaea	Zanthoxylum caribaeum	KJ012825.1		KJ082644.1					KJ426988.1						
Zanthoxylum	Zanthoxylum	Zanthoxylum flava	Zanthoxylum flavum	KJ012826.1		KJ082646.1					KJ426989.1						
Zanthoxylum	Zanthoxylum	Zanthoxylum martinicensis	Zanthoxylum martinicense	HM446754.2		HM446883.1					HM447014.1						
Zanthoxylum	Zanthoxylum	Zanthoxylum monophylla	Zanthoxylum monophyllum	KJ012827.1		KJ082647.1					KJ426990.1						
Outgroup-Sapindaceae	Outgroup for Fagara	Acer pseudoplatanus	Acer pseudoplatanus	KJ204427.1		HM849739.1					DQ978621.1						
Outgroup-Rutaceae	Outgroup for Fagara	Glycosmis pentaphylla	Glycosmis pentaphylla	AB762391.1		AB505903.1					JX856892.1						
Rhododendron	*Rhododendron*	*Rhododendron flavum*	*sy: Rhododendron luteum*	AY494182.1	AF072485.1				AY496923.1								
Rhododendron	*Rhododendron*	*Rhododendron hirsutum*	*Rhododendron hirsutum*	HE585263.1		HE585268.1											
Rhododendron	*Rhododendron*	*Rhododendron pontica*	*Rhododendron ponticum*	AY494172.1		KF997508.1			AY496913.1								
Outgroup-Ericaceae	*pyrola*	*Pyrola rotundifolia*	*Pyrola rotundifolia*	JN895849.1	FJ378588.1	JN893276.1			FJ378616.1								

Outgroup-Ericaceae	*leucopogon*	*Leucopogon lanceolatus*	*Leucopogon lanceolatus*	AY372642.1		JQ667345.1											
Hypericum	Hypericum	Hypericum androsaemum	Hypericum androsaemum	HQ331618.1	FJ694190.1	HQ332070.1											
Hypericum	Hypericum	Hypericum perforatum	Hypericum perforatum	JX661947.1	FJ694215.1	AF206779.1											
Hypericum	Hypericum	Hypericum quadrangulare	sy: Hypericum tetrapterum	JN894721.1	FJ694224.1	HQ332082.1											
Outgroup-Hypericaceae	Outgroup for Hypericum	Vismia cayennensis	Vismia cayennensis			JQ626022.1											
Outgroup-Clusiaceae	Outgroup for Hypericum	Symphonia globulifera	Symphonia globulifera	KC627885.1	AF479787.1	JQ625954.1											
Hibbertia	Hibbertia	Hibbertia cuneiformis	unresolved			FJ860367.1				FJ860484.1			FJ860427.1				
Hibbertia	Hibbertia	Hibbertia hypericoides	unresolved			FJ860377.1				FJ860494.1			FJ860437.1				
Hibbertia	Hibbertia	Hibbertia microphylla	unresolved			FJ860378.1				FJ860495.1			FJ860438.1				
Hibbertia	Hibbertia	Hibbertia salicifolia	sy: Hibbertia lucens			FJ860379.1				FJ860496.1			FJ860439.1				
Outgroup-Dilleniaceae	Outgroup for Hibbertia	Davilla rugosa	Davilla lacunosa			FJ860345.1				FJ860462.1			FJ860405.1				
Outgroup-Dilleniaceae	Outgroup for Hibbertia	Tetracera volubilis	Tetracera volubilis			KJ594530.1				FJ860512.1			FJ860456.1				

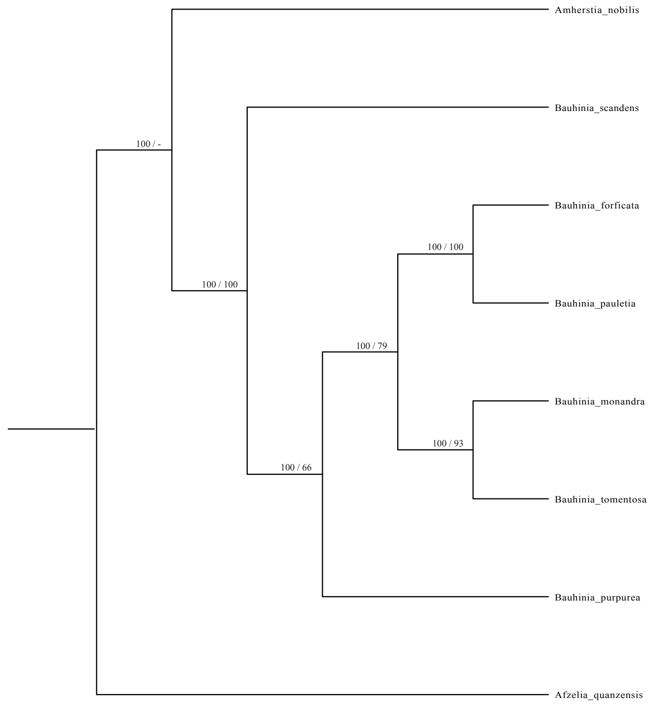
1. Dilleniaceae - Hibbertia:



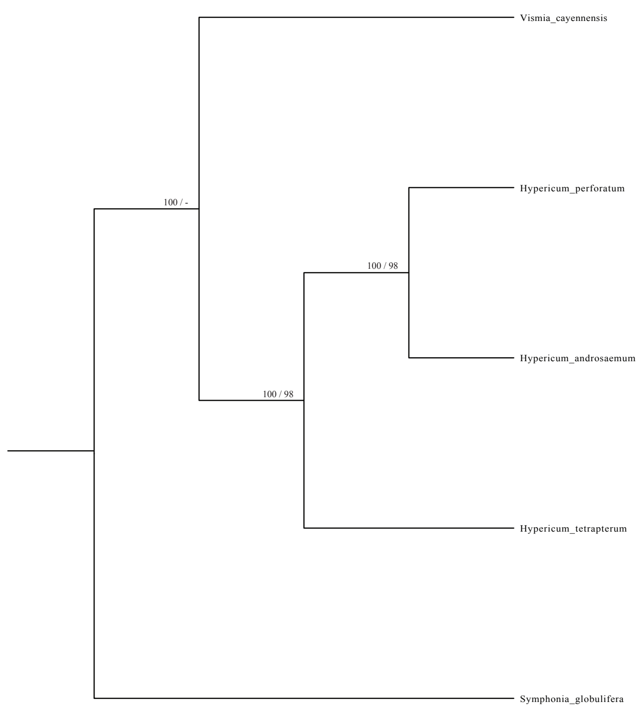
2. Ericaceae - Rhododendron:



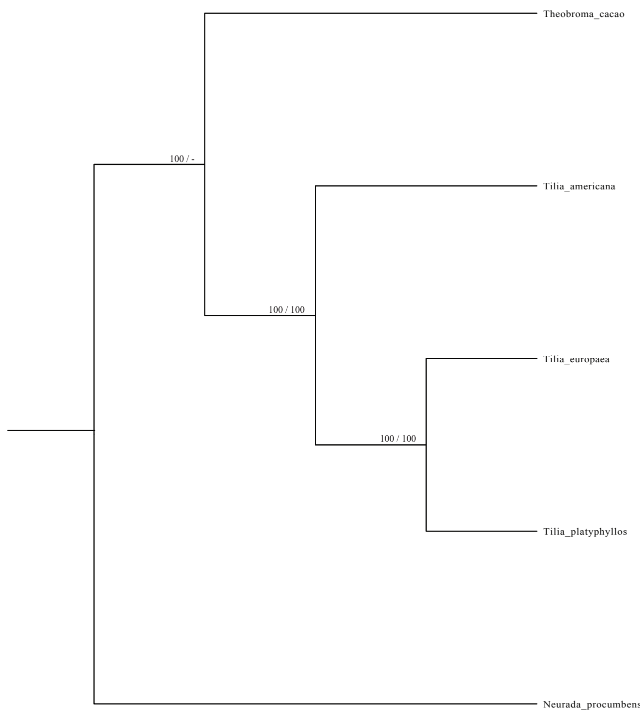
3. Fabaceae - Bauhinia:



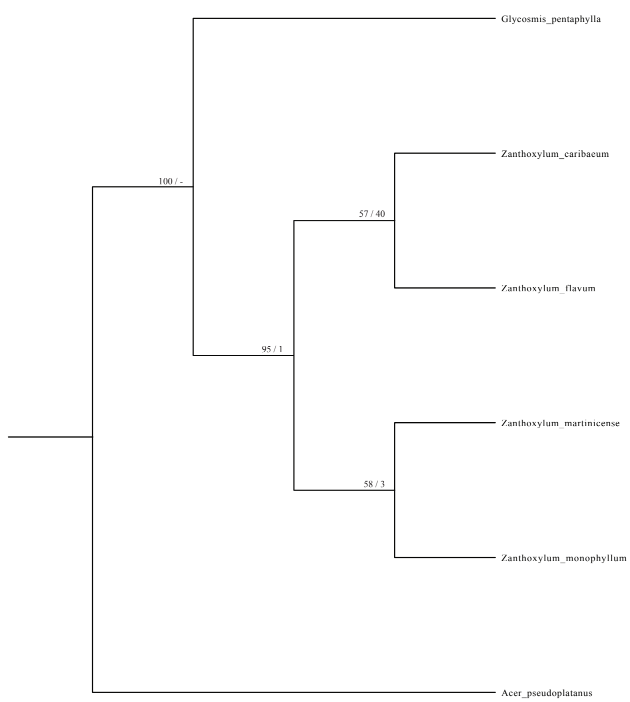
4. Hypericaceae - Hipericum:



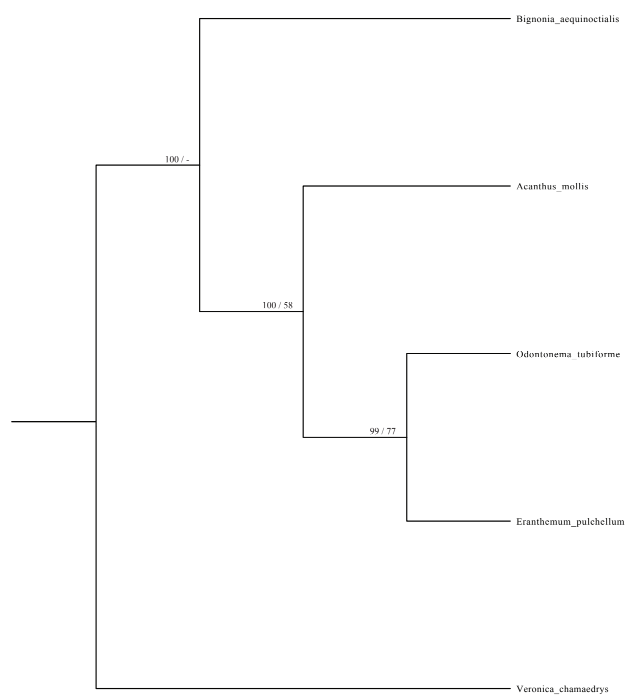
5. Malvaceae - Tilia:



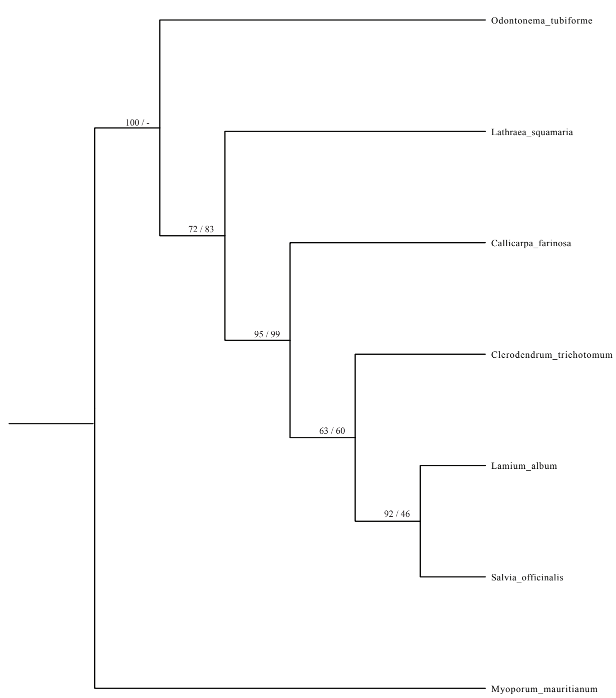
6. Rutaceae - Zanthoxylum:



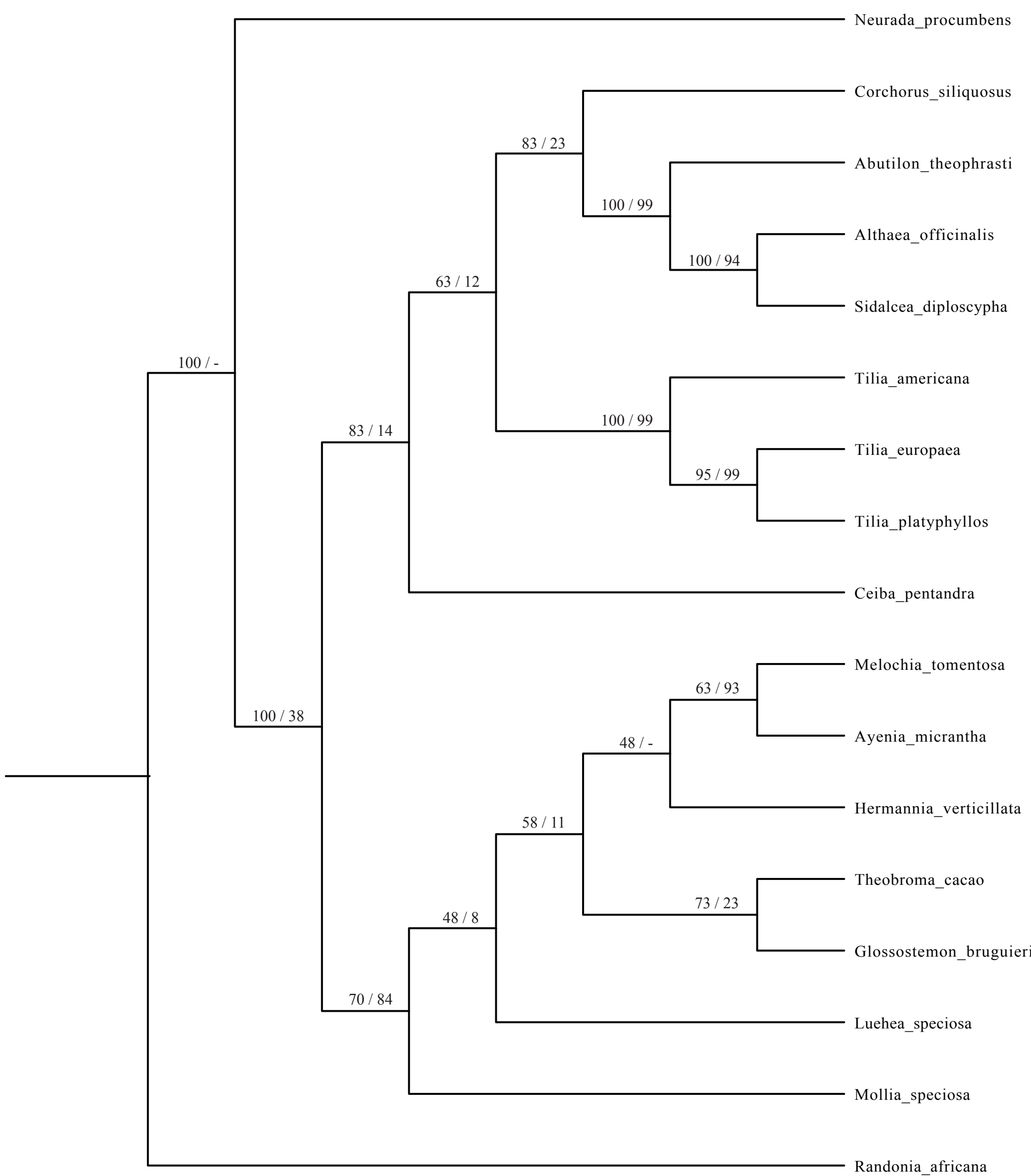
7. Lamiales - Acanthaceae:



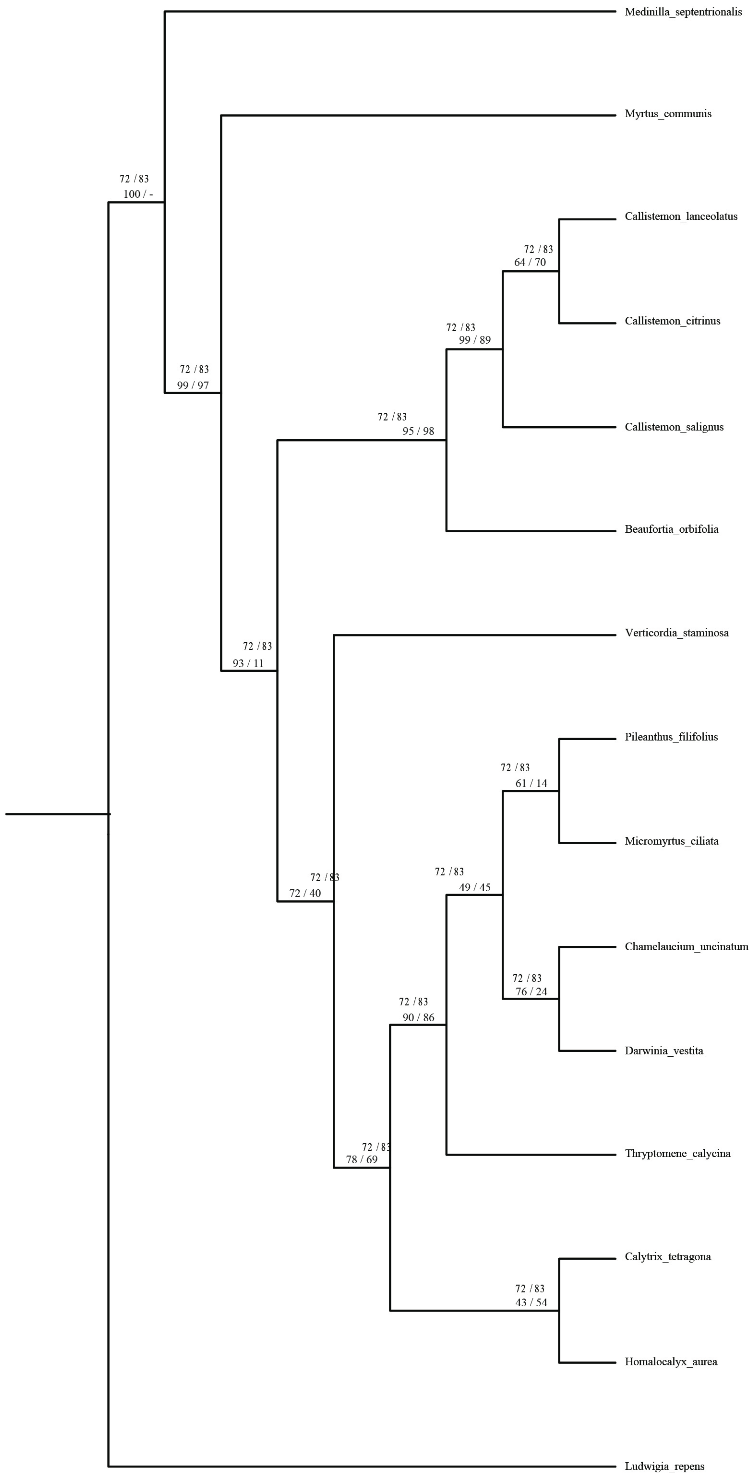
8. Lamiales - Lamiaceae:



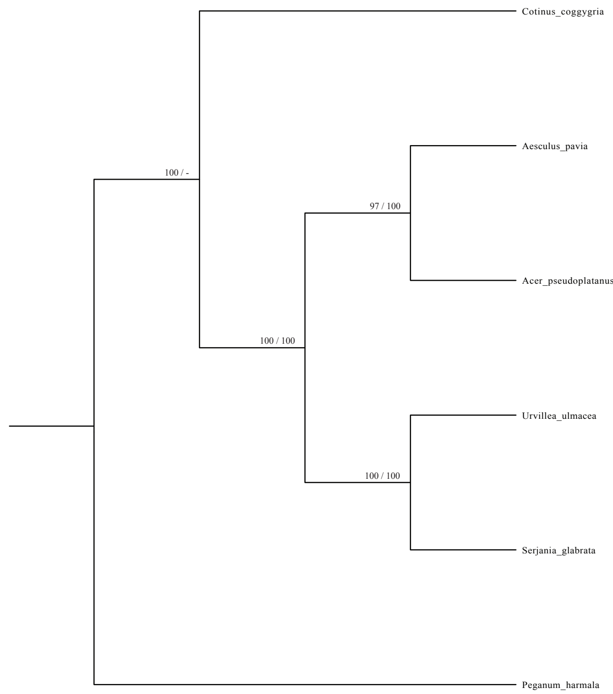
9. Malvales - Malvaceae:



10. Myrtales - Myrtaceae:



11. Sapindales - Sapindaceae:



Appendix figure 4. Phylogenetic reconstructions based on Bayesian and ML methods using molecular markers from Genebank. Phylogeny trees of 1. Dilleniaceae; 2. Ericaceae; 3. Fabaceae; 4. Hypericaceae; 5. Malvaceae; 6. Rutaceae; 7. Lamiales; 8. Lamiales; 9. Malvales; 10. Myrtales; 11. Sapindales. The phylogenetic relationships are based on the Bayesian approach. The ML trees show very similar topologies. The Bayesian posterior probabilities were show before the slash, while the ML bootstraps were show after the slash.

VITA

Ghadeer Bukhari was born on July 4th, 1987 in Jeddah, Saudi Arabia. Ghadeer received her Bachelor of Science in Zoology from the Biology Department with a Minor in Education from King Abdul Aziz University, Jeddah, Saudi Arabia in 2009. Soon after, she began an instructor position at King Abdul Aziz University in the Biology Department and received a scholarship allowing her to continue her higher education in the US. Ghadeer matriculated in the Master of Science graduate program in the Department of Biology at Virginia Commonwealth University (VCU) in 2014, and she is will begin PhD studies in the Integrative Life Sciences program in the Fall of 2016 at VCU.